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The main purpose of the Physiological Reviews is to furnish a means whereby those interested in the physiological sciences may keep in touch with contemporary research. The literature, as every worker knows, is so extensive and scattered that even the specialist may fail to maintain contact with the advance along different lines of his subject. The obvious method of meeting such a situation is to provide articles from time to time in which the more recent literature is compared and summarized. The abstract journals render valuable assistance by condensing and classifying the literature of individual papers, but their function does not extend to a comparative analysis of results and methods. Publications such as the Ergebnisse der Physiologie, the Harvey Lectures, etc., that attempt this latter task, have been so helpful as to encourage the belief that a further enlargement of such agencies will be welcomed by all workers. It is proposed, therefore, to establish a journal in which there will be published a series of short but comprehensive articles dealing with the recent literature in Physiology, using this term in a broad sense to include Bio-chemistry, Bio-physics, Experimental Pharmacology and Experimental Pathology.

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PHYSICO-CHEMICAL STUDIES ON BIOLUMINESCENCE

IV. The Physical and Chemical Nature of the Luciferase of Cypridina Hilgendorfii

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Harvey stated that the luminous secretion of Cypridina hilgendorfii gave no color reactions for proteins (2, p. 322). On the contrary, the writer observed that the luciferine and luciferase solutions are positive to the color reactions for proteins, though his publication in English was delayed over one year (7, p. 557). As the solutions just mentioned were by no means pure, it was not certain whether their color reactions were caused by the luminous substances themselves or some proteins present as impurities. Since then, therefore, the writer has been continuing the experiments to make clear the relation of protein reactions to the luminous substances. Recently, however, he found that Harvey has also published experimental results in the same line (5). But the writer's methods of experimentation and the results are not the same as Harvey's, as will be found in this paper.

The writer also conducted experiments on Cypridina luciferine so as to make comparisons with those of the luciferase. But the results of the former are not complete as yet. So the results of the latter experiments alone will be given in this paper.

The experiments were conducted in the Science Department of the Kyushu Imperial University, Fukuoka, Japan. The writer acknowledges his indebtedness to Professors Tsuneya Marusawa and Ayao Kuwaki for their interest and sympathy in the work. The work was supported by the private contribution of Mr. Jihachi Hamano.

Material. The preparation of the material was the same as already stated in the previous paper (8).

Methods. As the chief points of the methods were related to the solubility and precipitation of the luciferase, they will be stated in the following section.

Cypridina luciferase is soluble only in water. The writer, therefore, put the experimental material into distilled water in proportion of 1 gram to 50 cc., and stirred it thoroughly once in a while. Two hours later the material was filtered out and the clear filtrate was used for experiments. The material which had previously been washed by several changes of ether during the course of a few days might be considered fat-free. In the filtrate, however, there would be various substances, such as Cypridina luciferine, salts, some carbohydrates and proteins, which are all soluble in water. Only slight amounts of chlorides or of carbohydrate were present. On the other hand, the filtrate was positive to all color reactions, precipitation by "salting-out," by heavy metal salts and by alkaloidal reagents and heat coagulation characteristic for proteins which are soluble in water.

It should be considered, therefore, what kinds of proteins might be dissolved in the filtrate, when water was used as a solvent. Albumins are soluble in water and coagulable by heat. Globulins in general are coagulable by heat, but insoluble in salt-free water. As very little chloride was in the filtrate in question, it may be considered doubtful whether globulins were present. It should be remembered, however, that there are some globulins, called pseudo-globulins, in blood serum which are soluble in salt-free water. Such globulins, therefore, should be kept in consideration. Histone, proteoses, peptones and gelatins are soluble in water, but not coagulable by heat. At least, therefore, six groups of proteins might possibly be found in the filtrate.

A question arose as to what relation existed between the luciferase which is soluble in water and coagulable by heat and the proteins mentioned above as existing in the filtrate. The filtrate was nearly completely precipitated by alkaloidal reagents but poorly by heavy metal salts. It was also nearly completely precipitated by saturation with $MgSO_4$ and $(NH_4)_2SO_4$. Harvey, therefore, concluded that "only the group of albumins has properties which agree completely with those of luciferase" (5, p. 290); "but not a globulin" (5, p. 270). This conclusion of Harvey, based on the fact of "salting-out," seems to be too hasty. As a rule albumins are precipitated by saturation with $(NH_4)_2SO_4$ but not by saturation with $MgSO_4$ in neutral solution and at room temperature. On the other hand, globulins are precipitated by saturation with $MgSO_4$ and by half-saturation with $(NH_4)_2SO_4$. If so,

since luciferase is nearly completely or completely precipitated by saturation with either $MgSO_4$ or $(NH_4)_2SO_4$ it can hardly be determined by this test whether it belongs to the group of albumins or that of globulins. Furthermore, according to Osborne, certain vegetable globulins are not precipitated by $(NH_4)_2SO_4$ "until a concentration is reached greater than that secured by half-saturation" (6, p. 102). On the other hand, an albumin, the leucosin of the wheat germ, "is precipitated from its solution upon half-saturation with $(NH_4)_2SO_4$. The limits of precipitation by $(NH_4)_2SO_4$, therefore, do not furnish a sufficiently accurate basis for the differentiation of globulins from albumins. It has further been determined that a given protein which is precipitated by $(NH_4)_2SO_4$ cannot be 'salted-out' by the same concentration of the salt under all conditions" (6, p. 103). Thus one should be careful in attempting to decide the property of luciferase by "salting-out" precipitation.

Furthermore the luciferase is poorly precipitated by heavy metal salts especially by $HgCl_2$. Harvey also recognizes this fact (5, p. 286). If $HgCl_2$ was added to the filtrate in question, that is, the luciferase solution, a voluminous precipitate formed. Nevertheless a very little luciferase, which is assumed not to have been thoroughly washed out, was found in the precipitate. It is well known that albumins are well precipitated by $HgCl_2$. If, as Harvey states, the luciferase is an albumin, it should be precipitated by the salt. But this was not the case. Lead acetate, especially basic lead acetate, nearly completely precipitates the luciferase, but it is poorly precipitated by $AgNO_3$, $CuSO_4$ and $FeCl_3$.

From these facts just mentioned, the writer planned the following methods. As $HgCl_2$ precipitated out a voluminous substance from the luciferase solution but does not precipitate the luciferase, it was thought possible to separate the latter from the former, which was present as impurity. If so, the filtrate would contain the luciferase in comparatively pure state. If the filtrate was used for experiments, therefore, results would be more reliable than those when the luciferase solution which was not treated by $HgCl_2$ was used.

Proceeding on this theory 4 grams of the experimental material were put into 200 cc. of distilled water. It was stirred up once in a while, and after two hours, the material was carefully filtered. About 20 cc. of 2 per cent $HgCl_2$ were added to 150 cc. of the clear filtrate. A voluminous precipitate formed. The precipitate was filtered out with special care. The filtrate if not perfectly clear was returned to

the filter paper. This precaution was frequently necessary. The clear filtrate was again filtered through a heavy filter paper to make sure that no precipitate was left in it. No further precipitate formed when an excess of $HgCl_2$ solution was added to the clear filtrate. The luciferase remained in the filtrate intact. For convenience, the filtrate will be called the "experimental solution." The precipitate formed by addition of $HgCl_2$ solution was treated with water and the solution was positive to all color reactions for proteins except ninhydrin, although it was poorly dissolved in water.

The color reactions of the experimental solution. When Cypridina luciferine solution was added to the experimental solution in question, a brilliant light resulted, as Cypridina luciferase was in the latter. The luciferase was kept for many days with no perceptible injury. In other words, $HgCl_2$ seemed not to be "poisonous" to Cypridina luciferase.

The experimental solution was positive to the biuret, xanthoproteic, Millon, tryptophane and Molisch reactions. It was negative only to ninhydrin reaction. Most probably $HgCl_2$ interfered with this reaction. It is common that the reduced sulphur reaction forms a brown or black precipitate. In case of the experimental solution, the precipitate was gray. The Millon reaction forms a red precipitate or the solution becomes red when heated to 100°C.; but when the experimental solution was heated with the Millon reagent to 100°C., the solution showed no reaction. If, however, heating was stopped when the solution became slightly reddish, the reaction was positive. Chlorides interfere with the Millon reaction, and in this case it is necessary to add the reagent in excess. In case of the experimental solution, it was necessary to control heating besides the amount of the reagent. The writer tried in vain to determine whether the carbohydrate which gave the Molisch reaction was a starch or sugar. It might be a gum which was combined with protein. A carbohydrate radicle, such as glucosamine, is often present in the protein molecule. As before noted, this might be responsible for the Molisch test.

It is quite certain from the reactions mentioned above that some proteins still remained in the experimental solution. But it is not certain whether these proteins are the luciferase itself or have no relation to it.

Precipitation by alkaloidal reagents. When 0.5 cc. of 5 per cent phosphotungstic acid was added to 10 cc. of the experimental solution, an abundant precipitate formed. The precipitate was filtered out and partially dissolved in water. This solution produced a bright light

when mixed with the luciferine solution. But the clear filtrate, whether used as formed or diluted two or three times, produced no light when mixed with the luciferine solution. The luciferase was, therefore, completely precipitated by phosphotungstic acid.

When tannic acid and potassio-mercuric iodide were added to the experimental solution, an abundant precipitate formed, but the precipitate did not filter clear. So the results were not decisive. The writer thought that his failure was due to poor technique and tried these reagents in various ways in vain. When picric acid was added to the experimental solution, the solution became quite yellow, but no precipitate formed.

The experimental solution formed an abundant precipitate on addition of $K_4Fe(CN)_6$ plus acetic acid. This precipitate contained the luciferase and none was in the filtrate. The luciferase was, therefore, completely precipitated.

These facts favor the view that the luciferase is a protein, though they do not prove it; since, on the other hand, it may be said that the luciferase might be adsorbed by proteins which were precipitated by the alkaloids.

Precipitation by "salting-out." When the experimental solution was saturated with NaCl at 15°C., no precipitate formed. The solution filtered clear and the luciferase remained in the filtrate. Among proteins, there are some globulins which are precipitated by saturation with NaCl, but such seemed not to be present in the experimental solution.

When an equal volume of saturated $MgSO_4$ solution was added to the experimental solution, a slight precipitate formed. The solution filtered clear and luciferase was found to be abundant in the filtrate. The precipitate washed thoroughly with half-saturated $MgSO_4$ solution was dissolved in water and the solution produced a very faint light when mixed with luciferine solution. Half-saturation with $MgSO_4$, therefore, may precipitate some luciferase. Saturation with $MgSO_4$ produced a precipitate. This precipitate filtered clear and no luciferase was found in the clear filtrate The precipitate after being washed with saturated $MgSO_4$ solution was dissolved in water and the solution gave a bright light when mixed with luciferine solution This indicates that the luciferase was completely precipitated by saturation with $MgSO_4$. As already stated, proteins which are precipitated by saturation with $MgSO_4$ are generally globulins .

When an equal volume of saturated $(NH_4)_4SO_4$ was added to the experimental solution, a precipitate formed. This precipitate filtered

clear and no luciferase was found in the clear filtrate. The precipitate washed with half-saturated $(NH_4)_2SO_4$ solution was dissolved in water and the solution produced a bright light when mixed with luciferine solution. This means that half-saturation with $(NH_4)_2SO_3$ completely precipitated the luciferase. Proteins which are precipitated by half-saturation with the salt are globulins, primary proteoses and gelatin. It is hardly necessary to mention that saturation with $(NH_4)_2SO_4$ completely precipitated the luciferase, since half-saturation with the salt did so.

It may be worth mentioning that after the precipitates produced by saturation with $MgSO_4$ and by half-saturation with $(NH_4)_2SO_4$ were filtered out, both the clear filtrates gave color reactions for proteins except ninhydrin. It is possible that albumins, secondary proteoses and peptones which are not precipitated by such concentrations of these salts were left in the filtrates.

Coagulation by heat. When the experimental solution was heated at 55–60°C., it became slightly opaque, but the luciferase seemed not to be injured much. When it was heated at 65°C., it was coagulated, and the luciferase was entirely destroyed. It was not exactly determined at what temperature the solution was coagulated and the luciferase was destroyed. A mutual relation between the heat coagulation of the solution and the heat destruction of the luciferase seemed to exist. Proteins which are coagulable by heat are albumins, globulins and glutelins.

Precipitation by alcohol and acetone. By addition of acetone or alcohol to the experimental solution, an abundant precipitate formed. This precipitate filtered clear and no luciferase remained in the clear filtrate. The precipitate dissolved partially in water and the solution produced a brilliant light when mixed with luciferine solution. As the luciferase is only soluble in water, it is no wonder that the luciferase was precipitated entirely by acetone or alcohol. This precipitate dried thoroughly may be kept intact for a long time. But this precipitate formed by acetone or alcohol contains more impure substance than that formed by saturation with $MgSO_4$ or by half-saturation with $(NH_4)_2SO_4$. The precipitate formed by the former, however, is conveniently collected, but that formed by the latter is hard to collect.

Alcohol-soluble proteins are prolamins[1] and some substance found in Witte's peptone.[2] There is no protein found which is soluble in

[1] In Harvey's paper alcohol-soluble proteins are called protamines instead of prolamins (5, p. 281), but it may be a typographical mistake.

[2] This is not peptone but in reality largely a mixture of proteoses.

acetone. Alcohol and acetone, therefore, precipitate various groups of proteins and the precipitate is of course impure.

It may be well to summarize the experimental results hitherto mentioned in comparison with those of Harvey's in table 1. The blank in the table indicates no experiments made.

TABLE 1

Comparison of the experimental results of the physical and chemical nature of the luciferase of Cypridina hilgendorfii

KINDS OF EXPERIMENTS	HARVEY; LUCIFERASE SOLUTION; ONLY SOLUBLE IN WATER	KANDA, CLEAR FILTRATE, AFTER THE PRECIPITATE FROM LUCIFERASE SOLUTION FORMED BY $HgCl_2$ WAS FILTERED OUT; ONLY SOLUBLE IN WATER
Biuret reaction		+
Millon reaction		+
Xanthoproteic reaction		+
Tryptophane reaction		+
Molisch reaction		+
Ninhydrin reaction		−
Phosphotungstic acid	Completely precipitated	Completely precipitated
Tannic acid	Nearly completely precipitated	?
Picric acid	Nearly completely precipitated	?
$K_4Fe(CN)_6$ + acetic acid		Completely precipitated
Alcohol	Completely precipitated	Completely precipitated
Acetone	Completely precipitated	Completely precipitated
Saturation with NaCl	Not precipitated	Not precipitated
Half-saturation with $MgSO_4$	Not precipitated	Slightly precipitated?
Saturation with $MgSO_4$	Nearly completely precipitated	Completely precipitated
Half-saturation with $(NH_4)_2SO_4$	Slightly precipitated	Completely precipitated
Saturation with $(NH_4)_2SO_4$	Completely precipitated	Completely precipitated
Heat coagulation	Coagulable	Coagulable

Precipitation by "immune serum." The following experiments were planned to secure evidence as to whether Cypridina luciferine and luciferase are proteins. In all the following experiments, luciferase solution which was not treated by $HgCl_2$ was used. At the same time, luciferine solution was also used to make a comparison with the results of experiments on luciferase solution. If luciferine and luciferase are proteins, it was thought that the blood serum of an animal might be immunized after the preliminary injection of these solutions. That is to say, the serum of the injected animal might form a precipitate in luciferine or luciferase solution used to produce the immunity. If the serum of the animal injected with luciferase solution precipitates the luciferase in the solution, after the precipitate was filtered out, the filtrate might produce no light when mixed with luciferine solution. The same was thought with regard to luciferine solution. Each solution, therefore, was injected many times into a vein of a rabbit's ear. The serum of the rabbit was found to form a precipitate with luciferase or luciferine solution corresponding to the injected solution, as expected, though the amount of the precipitate was extremely small. And the desired experiments were performed in vain.

The failure of these experiments, however, does not prove that luciferase and luciferine are not proteins. Though the immune serum formed only a slight precipitate, there were quite a few groups of proteins in luciferase or luciferine solution. This means that the formation of the "immune serum" was not parallel with the presence of proteins in the solution and the failure seemed to be rather natural.

Dialysis and cataphoresis of the luciferase. Within forty hours after the treatment, the luciferase was not found outside of a parchment paper in which luciferase solution was placed. But at sixty hours after the treatment, a little luciferase which produced a very faint light when mixed with luciferine solution was found on the outside of the paper. This indicates that the luciferase solution might be of a colloidal state, but its particles would be so small that they could pass through the paper if kept running for a long time.

As some proteins and also electrolytes were in luciferase solution, there might be particles which were of different electrical charges in the solution. The writer tried to find out the sign of electrical charge of the luciferase. The right and left limbs of a U-shaped glass tube had one glass cock for each and at the bottom of the tube there was one three-way cock. About 5 cc. of luciferase solution were put in the tube and the right and left cocks were closed. The upper parts of the

limbs were carefully washed and about 1.4 cc. of distilled water was placed in each. Platinum wire electrodes were inserted into the distilled water of the limbs and were connected to the electrical current. Within ten minutes after this connection, some thick substance began to deposit at both cathode and anode. The substance deposited at the anode was not so thick as that at the cathode. About one hour after the treatment, substance accumulated at the anode reached to the three-way cock at the bottom of the U-tube. So it was made to run out through the cock and was received into a beaker. After this emptied part of the tube had been carefully washed with distilled water, the substance accumulated at the cathode was made to run out in the same way. Then luciferine solution was added to each of these substances run out. But neither produced light. In other words, the luciferase seemed to be destroyed by electricity. If so, it will be interesting to investigate the same with other enzymes, as there is no literature on this subject, as far as the writer is aware.

The same was tried with luciferine solution. The appearance of accumulated substances at both electrodes was about the same as that of luciferase solution. The substance accumulated at the cathode produced a bright light when mixed with luciferase solution, but the substance accumulated at anode produced no light. The luciferine, therefore, may be electro-positive. In this connection it is interesting to note that the luciferine is not precipitated by $K_4Fe(CN)_6$ plus acetic acid which is also electro-positive.

The author also tried to see whether any change of a luminous mixture of luciferine and luciferase solutions occurred when a current was sent through it. The result was that luminescence at both poles disappeared as substances were accumulated there. As the luciferase was destroyed by cataphoresis, as already stated, this seemed to be a natural result.

Electrical conductivity The writer tried to see whether the electrical conductivity was changed when light was produced on addition of a luciferase solution to a luciferine solution. But there was no change of electrical conductivity when light was produced and even after twelve hours. According to Harvey (4, p. 139), Dubois believes that Pholas luciferine, a protein, forms amino-acids by light production. If so, there should be a change of electrical conductivity in light production, as Bayliss' study on trypsin has shown (1).

DISCUSSION

From the results of "salting-out," Harvey concludes that Cypridina luciferase is not a globulin, but it may be an albumin. Judging from his results of "salting-out," however, it is neither an albumin nor a globulin, because it is "nearly completely precipitated" by saturation with $MgSO_4$ and is "completely precipitated" by saturation with $(NH_4)_2SO_4$, but not by half-saturation with the latter salt. Proteins which are precipitated by saturation with $MgSO_4$ are generally globulins, but not albumins; and those which are precipitated by saturation with $(NH_4)_2SO_4$ are albumins, but globulins are precipitated by half-saturation with $(NH_4)_2SO_4$. It is, therefore, hardly conclusive from Harvey's results that the luciferase is either an albumin or a globulin. In the writer's experiments of "salting-out," the luciferase is completely precipitated by saturation with $MgSO_4$ and by half-saturation with $(NH_4)_2SO_4$. From these results, therefore, it should be said that the luciferase is a globulin. But attention is called to the fact that globulins in general are not soluble in salt-free water. The writer first prepared the luciferase solution by dissolving the material in distilled water. A little chloride was in the solution, but it was doubtful whether that amount of chloride was sufficient to dissolve globulins in water. But on the other hand, there are some globulins in blood serum which are soluble in salt-free water. The possibility of the luciferase as a globulin, therefore, may not be entirely excluded by its solubility in nearly salt-free water. And the leucosin of the wheat germ, an albumin, is also "precipitated from its solution upon half-saturation with $(NH_4)_2SO_4$." Thus precipitation by "salting-out" is not an altogether accurate basis to differentiate globulins from albumins.

Although primary proteoses and gelatin are precipitated by half-saturation with $(NH_4)_2SO_4$, they are not heat-coagulable. They seem, therefore, to be quite different substances from the heat-coagulable luciferase. Histones are also soluble in water but not heat-coagulable. On the other hand, glutelins are heat-coagulable but insoluble in water, while the luciferase is soluble only in water. As water-soluble peptones are not heat-coagulable and are not precipitated by half-saturation with $(NH_4)_2SO_4$, they seem not to be related to the luciferase.

Thus it is possible that the luciferase may finally be determined to be an albumin, or a globulin which is assumed to be soluble in water containing very little chlorides or is assumed to be similar to the pseudo-globulin of blood serum. Although the luciferase is assumed to be

either an albumin or a globulin, it is specific in character and the protein character is not its whole nature, because the luciferase is not replaced by blood serum which contains a mixture of albumins and globulins. The question is, therefore, whether the reaction of the "experimental solution" as a protein is really a proper reaction of the luciferase or not. Moreover the "experimental solution" gave a carbohydrate reaction as well as the protein one. It is hard, therefore, to decide which is the reaction of the luciferase. In short, the question is how pure luciferase is obtained.

This is the question of enzymes in general. Amylases, for example, are regarded by some investigators, (Osborne and Wroblewski), as proteins but not carbohydrates, while by others[3] as carbohydrates but not proteins. It is hard to decide whether they are proteins or carbohydrates, although it is certain that they contain nitrogen in their composition. It is said that the part of enzymes which give a carbohydrate reaction is a gum. The luciferase may be, therefore, a sort of protein which is combined with a carbohydrate gum.

SUMMARY

1. The writer was the first to point out that the luciferine and luciferase solutions of Cypridina hilgendorfii are positive to color reactions for proteins. While he was continuing further investigation in that line, Harvey also published his results in the same line.

2. The luciferase of Cypridina is not precipitated by $HgCl_2$. The writer therefore, used for these experiments the clear filtrate after the precipitate formed by $HgCl_2$ was filtered out. This filtrate is called the "experimental solution."

3. The experimental solution is positive to the biuret, xanthoproteic, Millon, tryptophane and Molisch reactions but negative to ninhydrin.

4. Phosphotungstic acid and $K_4F_4(CN)_6$ plus acetic acid completely precipitate the luciferase, but precipitation by other alkaloidal reagents is not successful.

5. The luciferase is not precipitated by saturation with NaCl. But it seemed to be more or less precipitated by half-saturation with $MgSO_4$. It is completely precipitated by saturation with $MgSO_4$ and by half-saturation with $(NH_4)_2SO_4$. After the precipitate formed by these

[3] Fraenkel and Hamburg found that very active preparations of amylases "gave no protein reactions, except the faintest Millon and xanthoproteic. The carbohydrate reaction was strong" (9, p. 330).

salts is filtered out, the clear filtrate is still positive to the biuret, xanthoproteic and Millon reactions but not to ninhydrin.

6. The luciferase is coagulated at about 65°C.

7. The luciferase is completely precipitated by acetone and alcohol.

8. The precipitation of the luciferine and luciferase by "immune serum" is not successful.

9. The luciferase seems to pass through a parchment paper, after sixty hours treatment, in slight amount.

10. The luciferase is completely destroyed by cataphoresis. But the luciferine seems to be electro-positive. A light-producing solution gradually loses its light as a precipitate is formed by cataphoresis.

11. Light production does not change electrical conductivity.

12. Judging from the results mentioned above, the luciferase may be colloidal and a globulin. A carbohydrate gum may be combined with a protein as a part. It is not evident, however, whether the luciferase is really a protein or a carbohydrate or a substance which has both natures. The luciferase may be absorbed by a protein. At any rate, the question is not solved unless the luciferase is identified with a substance which gives a protein reaction.

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STUDIES ON THE VISCERAL SENSORY NERVOUS SYSTEM

IV. The Action of Certain Drugs on the Lung Motor Mechanisms of the Reptilia (Turtle, Snake)

A. B. LUCKHARDT and A. J. CARLSON

From the Hull Physiological Laboratory of the University of Chicago

Received for publication August 23, 1920

These observations on drug action on the lungs were made primarily for the purpose of further elucidation of the lung motor control through the extrinsic nerves. The use of curare was incidental in the preparation of some of the animals for experimentation.

The reader is referred to our previous paper on lung automatism and lung reflexes in the reptilia for description of methods of preparing the animals and recording the lung contraction (1).

The literature on the action of drugs on the lung motor mechanism of reptilia is meager. Francois-Franck (2) and Prevost and Saloz (3) state that atropine paralyzes the motor fibers in the vagi nerves. Jackson and Pelz (4) state: "After atropine we have sometimes seen what undoubtedly appeared to be direct muscular contractions." This may mean that atropine itself caused lung contractions; but the statement is not clear nor is it exemplified by the tracings secured.

Prevost and Saloz also report that atropine abolishes or prevents the prolonged lung contractions caused by pilocarpin and muscarin, while in preparations not atropinized vagus stimulation augments the motor action of these alkaloids on the lungs. Jackson and Pelz found that nicotin and pituitrin cause prolonged lung contractions.

The studies of Jackson and Pelz on the adrenalin action in the turtle's lung were inconclusive. Typical lung dilatation by adrenalin was not obtained, but the authors suggest that this may have been due to faulty technique. In Jackson's method of preparing the turtle for recording the lung contractions, there is greatly impaired circulation not only through the lungs but in the rest of the body.

Heinekamp (7) has recently reported evidence tending to show adrenalin has a direct stimulating action on the cardiac vagus center in

the turtle. In his experiments the spinal cord was cut in the neck and pithed peripherally. The head was excluded from the peripheral circulation, and the drug injected through the central end of one carotid. Such a preparation passes gradually into asphyxia, and in the turtle asphyxia causes cardiac inhibition both by central (medulla) and peripheral (direct on the heart) action. While this is a serious source of error, the fact that Heinekamp obtained cardiac inhibition when perfusing the medulla with adrenalin Ringer solution, but not with the Ringer solution alone, points to the probable correctness of his conclusion.

I. RESULTS ON THE TURTLE (CHRYSEMYS ELEGANS AND MALACOCLEMMYS LESUEURII)

Curare. Intravenous injection of curare in the turtle abolishes or depresses the lung motor mechanism for periods of a few minutes to several hours (fig. 1, *A*). This depression is most marked in animals in poor condition. Parallel with the depression of the lung rhythm there is a very pronounced cardiac inhibition, probably by central vagal stimulation. This curare effect on the heart is more temporary than the depressor action on the lung rhythm. Curare also depresses gastric motility (the peripheral automatism) over even longer periods than its depressor action on the lung motor mechanism (fig. 4).

The point of action of this drug in depressing the lung contractions was not determined completely. It is well known that curare has a primarily stimulating action on the central nervous system. This is always observed in the turtles in good condition. Turtles in poor condition, on the other hand, go into the typical curare paralysis without evidence of primary central stimulation (general restlessness and struggling).

Paralysis of the skeletal musculature reduces the total CO_2 production in the body, and since the lung contractions are due to discharges from the respiratory center into the lung motor center of the vagus, the curare depression of the lung rhythm may be due to this lessened CO_2 production and consequent slower and feebler action of the respiratory center. We do not believe that this is the essential factor, because curare will depress the lung motor mechanism even in preparations already completely curarized. In this case there can be no diminution in CO_2 production by skeletal muscle paralysis, unless we assume a direct depressor action of curare on the muscle cells.

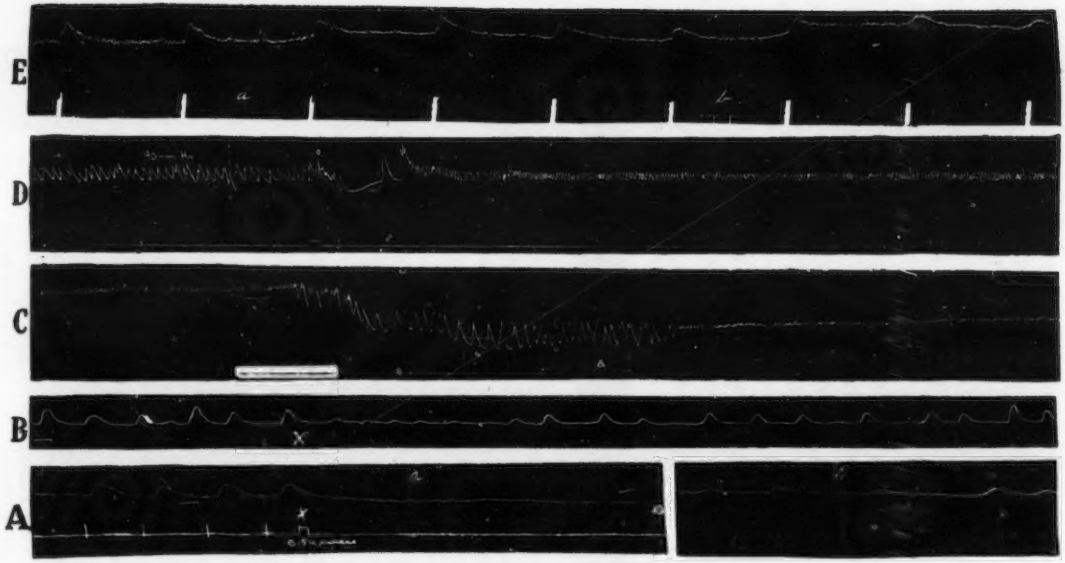

Fig. 1. Turtle; decerebrated. Fixed on stand ventral side down. Left lung isolated except for pulmonary vagi and blood vessels. Cannula in tip of left lung, left bronchus ligated.

A and *B*, water manometer tracings of lung contractions; *a*, animal not curarized. Signal, spontaneous respiration; *b*, section of tracing 20 minutes later than *a*; *x*, intravenous injection of 0.5 cc. of curare in Ringer's solution. *B*, animal curarized prior to the curare injection at *x*. Showing depression of the lung motor rhythm by curare.

C and *D*, carotid blood pressure. Signal, intravenous injection of 0.5 cc. 1 per cent curare in Ringer's solution. Showing prolonged vagal stimulation by curare.

E, Isolated left lung, left vagus sectioned in neck, animal completely curarized. Signal, tetanization of vagus for 5 seconds every 2 minutes. Intravenous injections of curare *a*, 0.5 cc.; *b*, 1.0 cc. 1 per cent curare. Showing very slight peripheral lung depression by large doses of curare.[1]

[1] All figures have been reduced two-thirds to three-fourths of the original.

When the peripheral vagus is tetanized for short periods at regular intervals contractions of the corresponding lung are obtained, of surprising regularity in strength and duration. Intravenous injection of large doses of curare under such conditions causes only a slight depression of the lung contractions (fig. 1, *E*).

The interference of curare with the lung motor mechanism is therefore mainly central, or a depression of the respiratory center itself. But in case of the empty stomach the peripheral depressor action of this drug is very marked. In preparations with the medulla and vagi intact, and breathing spontaneously, curare in quantities from 0.2 to 0.5 cc. of 1 per cent will inhibit the contractions of the empty stomach for hours.

Picrotoxin. In our previous communication it was shown that the rhythmic lung contractions are essentially of central origin (1), a side event of the respiratory discharge in the medulla. We might there-

Fig. 2. Water manometer record of the intrapulmonic pressure of the turtle's lung. The tracings show reflex contractions of the isolated left lung from a single inflation of the right intact lung,; *a*, before and after (*b* = 15 minutes; *c* = 30 minutes) the intravenous injection of 0.1 mgm. picrotoxin.

fore expect medullary stimulants such as picrotoxin and caffeine to augment the lung contractions. Our results with citrated caffeine were inconclusive. Intravenous injection of this drug even in large doses (10 mgm.) does not initiate a lung rhythm in quiescent preparations, nor does it appreciably augment the rhythm in active preparations; or if there is any stimulating action by this drug, it is so slow and gradual that it becomes impossible to differentiate it from other factors such as spontaneous fluctuations in the rhythm or the effects of slight asphyxia.

Picrotoxin, on the other hand, has a distinct stimulating action on the lung motor mechanism. This action is central, the drug being unable to initiate a lung rhythm after section of the vagi. Picrotoxin accelerates the rhythm in active preparations, showing particular tendency to induce the incomplete lung tetanus characteristic of marked asphyxia. In quiescent preparations intravenous injection of picrotoxin increases the reflex excitability of the lung motor mechanism

Fig. 3. Water manometer tracing of the intrapulmonic pressure in the turtle. Animal decerebrated and completely curarized. Record from left lung, isolated except for pulmonary vagi and blood vessels. Shows spontaneous lung rhythm. Intravenous injections of 0.3 mgm., *a*, 0.6 mgm., *b*, picrotoxin. *c*, stimulation of central end of right vagus. Showing augmentation of the lung rhythm and the reflex excitability of the lung motor center by picrotoxin.

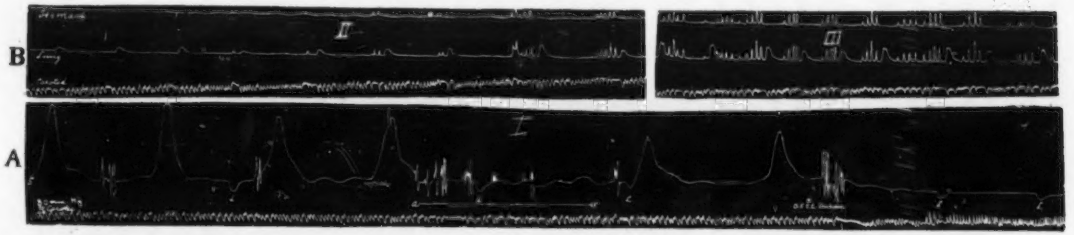

Fig. 4. Records of gastric contractions (empty stomach, balloon method), lung contractions and carotid blood pressure (tambour) in the decerebrated turtle. There is a 10-minute interval between end of tracing *I* and beginning of tracing *II;* and a 16-minute interval between tracings *II* and *III*. *I, S,* stomach record; *B,* carotid blood pressure; *L,* lung contractions shown as waves of negative pressure on the stomach record; *a-a'*, isolation of external jugular vein; *x*, intravenous injection 0.5 cc. 1 per cent curare in 5 cc. Ringer. *II* and *III* upper record, stomach; middle record, lungs (from cannula in trachea, water manometer); lower record, carotid blood pressure.

Showing prompt inhibition of the strong stomach contractions and gradual depression of the feeble and more rapid stomach tonus rhythm, while the respiratory mechanisms (external respiration and lung contractions) suffer initial depression and show gradual recovery.

In *II* and *III* the quick upstrokes = external respiratory movements (also appearing on the stomach record); the slower lung contractions following at the end of the external respiration do not appear on the stomach record.

before or without initiating a rhythm. Typical tracings illustrating the picrotoxin action on the lung rhythm and reflexes are reproduced in figures 2 and 3.

Nicotine. Small quantities of nicotine (0.05 to 0.1 mgm.) injected intravenously in the turtle cause a temporary inhibition of the lung motor mechanism (fig. 5, *A*). This inhibition is evidently central, for with these doses no peripheral paralysis or depression of the lung motor mechanism can be demonstrated.

In larger concentrations nicotine causes contractions of the lung musculature, evidently by direct peripheral action, as they occur after section of the vagi. These contractions are not due to stimulation of the vagi nerve endings in the lung, because on repeated injections of nicotine the drug soon ceases to produce lung contraction, while the vagi motor fibers are still active on the lung (fig. 5, *B*).

It is not possible to paralyze the pulmonary motor fibers by nicotine except in quantities that act as a general protoplasmic poison. In this respect the vagi lung motor fibers in the frog are similar to those of the turtle, both being resistant to nicotine paralysis. We do not believe that this fact warrants the conclusion that there are no ganglion cells interposed in the path of the motor pulmonary fibers. The lung motor fibers in the frog and turtle are similar to the cardio-accelerator nerves in their resistance to nicotine but differ from the latter in their great susceptibility to the paralyzing action of atropin.

There is also a difference in the vagi-lung motor fibers and the vagi-gastric motor fibers in their resistance to nicotine. Working in this laboratory, Bercovitz and Rogers have recently found that relatively large doses of nicotine paralyze the vagi-gastric motor fibers after a prior depression of the spontaneous contractions of the empty stomach, but on stimulation of the vagi after these large doses of nicotine a temporary inhibition of the gastric tonus is produced, indicating that the vagi-gastric inhibitory fibers are still capable of functioning.

On the basis of our results on the amphibia we might expect that the possible pulmonary inhibitory nerves would be paralyzed by doses of atropin that leave the motor nervous mechanism intact. But apart from the effects noted above nicotine causes no changes in the spontaneous lung contraction or in the motor response to vagal stimulation that might indicate the presence of inhibitory nervous mechanism to the lungs in these responses in the normal animal.

At no time did we note any direct inhibitory action of nicotine on the turtle lung similar to that found in frogs and salamanders.

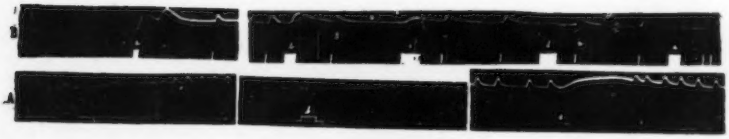

Fig. 5. Tambour tracings of the intrapulmonic pressure in the turtle. Record from left lung, isolated except for pulmonary vagi and blood vessels. Left bronchus ligated. Cannula in tip of lung. Animal decerebrated and partially curarized.

A, spontaneous lung rhythm; *a*, *b* and *c*, successive intravenous injections of 0.1 mgm. nicotine in Ringer's solution, showing temporary inhibition (central) of the lung contractions.

B, left vagus sectioned in the neck and corresponding lung quiescent. Signal, tetanization of left peripheral vagus for five seconds. Letters, intravenous injections: *a*, 1 mgm. nicotine; *b*, 1 mgm. nicotine; *c*, 5 mgm. nicotine; *d*, 5 mgm. nicotine; *e*, 0.5 mgm. atropine sulphate. Showing resistance of pulmonary vagus to nicotine, and the prompt paralysis by atropin (the heart beat is shown on these lung tracings).

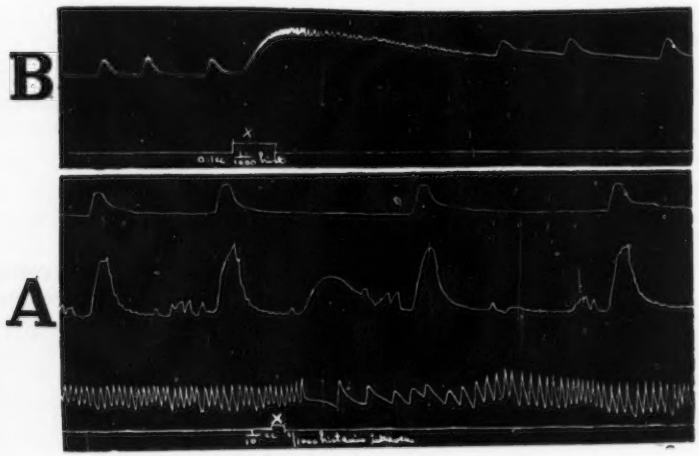

Fig. 6. *A*, water manometer records of the rhythmical contractions of the turtle lung. Animals decerebrated, partially curarized and fixed on stand ventral side down. Left lung (upper tracing) isolated except for vagi and blood vessels. The latter occluded by gentle traction on lifting ligature about them. Middle tracing from right intact lung. Lower tracing, blood pressure tracing from right carotid artery.

X = intravenous injection through the external jugular vein 0.1 cc. 1/1000 histamine-HCl solution.

Showing peripheral action of histamine on the lung (middle tracing), absence of any central action (upper tracing), and marked cardiac inhibition.

B, tambour record of isolated left lung as in *A*. *X*, intravenous injection of 0.1 cc. 1/1000 histamine. Showing temporary central action of histamine (slowing of the medullary rhythm). The heart beat appears on part of tracing.

Histamine. Histamine hydrochloride causes contractions of the turtle lung. In weak concentrations the drug induces a single lung contraction only slightly more prolonged than the contractions in the normal lung rhythm. Stronger concentrations cause prolonged tonus or tetanus contractions. The contractions caused by weak concentrations of histamine may be considerably weaker than those of the normal lung rhythm.

The histamine appears to cause lung contractions by direct actior on the lung musculature, because they occur after atropin paralysis of the vagi motor fibers (figs. 7, 8). The histamine lung contractions are not due to motor discharges from the medulla, as they occur after section of the vagi, but they do not appear if previous to the intravenous injection of the drug the pulmonary blood vessels have been ligated (fig. 6). The lung continues to respond for hours to the central motor discharges via the vagi after occlusion of the pulmonary artery and vein.

In several turtle preparations intravenous injections of histamine were made while records were being taken of the lung as well as of the stomach contractions (balloon method). It was noted that histamine concentrations causing very strong lung contractions failed to induce contractions of the gastric musculature (fig. 7, *A*). In the turtle at least the lung musculature is very much more sensitive to histamine than is the gastric musculature.

When histamine is injected intravenously in preparations showing the normal lung rhythm there appears a temporary depression or inhibition of the lung motor center. This inhibition may, indeed, be indirect or through depression of the respiratory center, and the depression of the latter may be in part a reflex effect, afferent impulses being set up by peripheral smooth muscle contractions.

The stimulating action of histamine on the lung musculature as well as its depressor action on the medulla decreases rapidly on repeated intravenous injections of the drug. It does not seem possible to maintain the lung musculature in a continued state of hypertonus or incomplete tetanus by repeated intravenous injections.

The reader will recall that histamine has a primary inhibitory action on the lung motor tissues of the salamanders (necturus and axolotl). This primary inhibitory action by histamine was never observed in the lung of the turtle. The reason for this difference lies probably in the predominance of the inhibitory nervous mechanism in the salamander, the histamine stimulation of the latter counteracting or

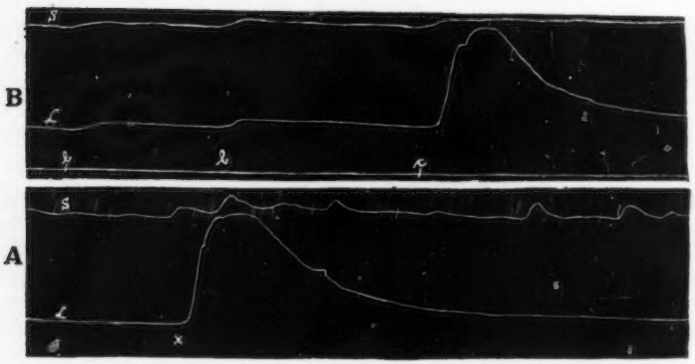

Fig. 7. Water manometer tracing of the intrapulmonic pressure in the turtle. Animal decerebrated and fixed on the stand ventral side down and curarized. Cannula in tip of left lung, isolated except for the blood supply. Left bronchus ligated. Left vagus sectioned in neck paralyzing left lung. *L*, record from lung; *S*, record from balloon in empty stomach.

A at *X* = injection of 0.2 cc. 1/1000 histamine intravenously.

B at *b* = two intravenous injections of ½ mgm. each of atropin sulphate *c* = intravenous injection of 0.2 cc. 1/1000 histamine.

Showing the peripheral and probably direct action of histamine on the lung musculature after paralysis of vagus nerve endings with atropin, and feeble action, if any, on the gastric musculature of histamine in this concentration.

Fig. 8. Water manoneter record of the intrapulmonic pressure in the left isolated lung of a decerebrated and partially curarized turtle. Left vagus sectioned in neck. Cannula in tip of left lung. Left bronchus ligated.

a = stimulation of left peripheral vagus every 2 minutes for 5 seconds with weak tetanizing current.

X = Intravenous injection of 0.5 cc. 0.1 per cent atropin sulphate.

↓ = indicates intravenous injection of 0.2 cc. 1/1000 histamine hydrochloride solution.

Showing the paralyzing action of atropin on the terminations of the vagus nerve as well as the peripheral action of histamine on the lung musculature after atropinization.

obscuring its direct muscular action. If this interpretation is correct, we might expect a histamine stimulation of the lung motor nervous mechanism in animals like the turtle in which this is predominating while the inhibitory nervous mechanism is feeble or absent entirely. In this case paralysis of the lung motor fibers by atropin should weaken the lung contractions induced by moderate doses of histamine due to exclusion of one of the points of stimulation. We have been unable to determine whether this is a fact, mainly because two successive injections of the same dose of the drug rarely induce lung contractions of equal amplitude and duration, due to the gradually decreasing efficiency of the drug on repeated injections.

Atropin. Atropin in small doses (0.5 mgm. or less) promptly paralyzes the vagi motor fibers to the lungs. This has been recorded by a number of investigators. In fact, after atropin, stimulation of the peripheral vagus has no effect either in the direction of motor or inhibitory effects. In this respect there is a striking difference between the reptilian and the amphibian lung motor mechanism, as in the latter group of animals atropin, even in enormous doses, has little or no effect either on the inhibitory or on the motor nervous mechanism controlling the lung musculature.

The great resistance of the turtle to the general poisonous action of these drugs is noteworthy. One small turtle weighing only 625 grams and prepared in the usual way by decerebration and isolation of the left lung, the preparation breathing spontaneously by the intact right lung, received during the period of seven hours twelve intravenous injections of adrenalin (total: 4.15 cc. 1/1000); six injections of nicotine (total: 2.2 mgm.); five injections of histamine (total: 1.35 cc. 1/1000); 1 cc. Armour's pituitary liquid. At the end of this period the heart, respiration and lung rhythm were distinctly weaker than normal; but, left over night, the preparation was in nearly normal vigor the following morning. This is evidence of great drug tolerance as compared to the mammal, as so small a dose as 0.5 cc. 1/1000 histamine intravenously being able to kill a 10-kilo dog.

Adrenalin. If adrenalin in a dose of 0.1 cc. 1/1000 or less is injected intravenously in a decerebrated animal, normal in every respect save for a tracheotomy from which a record of external respiration is being obtained, the immediate effect is a depression of the amplitude of the external respiratory movements. This is shown in figure 9, *A*, where the fairly rapid and deep respiratory movements of the Cheyne-Stokes type were almost immediately changed, following the injection of the

adrenalin at *X*, to respiratory groupings which were less vigorous in character.

The left lung was now isolated in the usual manner. Its blood supply and innervation through the vagus were conserved. Record of the lung rhythm was obtained from the tip of the isolated lung, the left bronchus having been tied off. A record of the normal lung rhythm having been

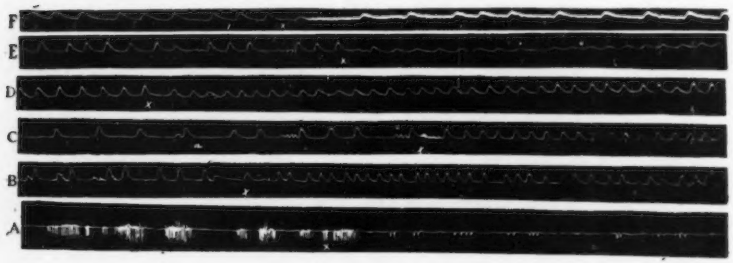

Fig. 9. Tambour record of the intrapulmonic pressure in the turtle. Animal decerebrated and fixed on stand ventral side down. All injections into jugular vein, in 5 cc. Ringer's solution.

A, shell intact, record from side tube of tracheal cannula. Spontaneous respiration. *X*, intravenous injection of 0.1 cc. 1/1000 adrenalin, showing weakening of the external respiratory movements.

B, record from the left lung, isolated except for pulmonary vagi and blood vessels. Cannula in tip of lung; left bronchus ligated. Right lung intact; animal breathing spontaneously. These tracings show the lung muscle rhythm.

B, *x*, injection of 0.05 cc. 1/1000 adrenalin.

C, *a*, injection of 5 cc. Ringer solution; *x*, injection of 0.1 cc. 1/1000 adrenalin.

D, *x*, injection of 0.5 cc. 1/1000 adrenalin.

E, *x*, injection of 1.0 cc. 1/1000 adrenalin.

F, same preparation 12 hours later; *x*, injection of 1.0 cc. 1/1000 adrenalin.

Showing acceleration of the lung rhythm by small, and depression (weakening and slowing) by large quantities of adrenalin.

obtained (fig. 9, *B*) 0.05 cc. of 1/1000 adrenalin solution was injected at *X*. It is clear that the effect of this intravenous injection was an increase in the rate and diminution of the amplitude of the lung contractions. This increase in rate was not due to an increased discharge from the lung motor center because of an improved circulation through it, as the animal was given an intravenous injection of an equivalent amount of Ringer's solution at *a*, figure 9, *C*. This had no effect; but the subsequent injection at *X* of 0.1 cc. of 1/1000 adrenalin caused

again an accelerated lung rhythm. The rate was virtually doubled. Furthermore, increased circulation through the medulla because of the hemodynamic effects of adrenalin should have slowed rather than accelerated the respiratory rhythm.

The injection of larger doses diminished the amplitude of the contraction without appreciably affecting the rate. These points are well shown in figure 9, *D*, and figure 9, *E*, where the animal received intravenous injections of 0.5 cc. and 1.0 cc. respectively of 1/1000 adrenalin solution. In both instances these larger doses diminished the vigor of the rhythm without appreciably affecting the rate. If anything the rate was here, too, slightly increased.

The injection of a large dose (1 cc. 1/1000 adrenalin) some twelve hours later not only reduced the rate of the rhythm but also diminished the amplitude of the individual contractions, figure 9, *F*.

The cause of the depression of the lung motor activity, under certain conditions, as the result of intravenous injection of adrenalin engaged our attention for some time. The temporary depression of rate and amplitude of the spontaneous lung contractions following the intravenous injection of 0.1 cc. of 1/1000 adrenalin solution into an animal partly asphyxiated because of loss of blood is very strikingly shown in figure 10, *A*. It is not obvious, however, whether the general depression of lung motor activity is due to peripheral or to central action of the drug, although diminution in rate itself strongly indicates central action.

The experiment was repeated with this difference that the blood vessels of the isolated lung were occluded by traction on a lifting ligature placed about them preceding the intravenous injection of 0.2 cc. 1/1000 adrenalin solution. Since the depression of lung motor activity was almost identical in both lungs (fig. 10, *B*) in spite of the fact that the adrenalin was prevented from reaching the pulmonary tissue of the isolated lung, we conclude that adrenalin acts essentially by depresing the lung motor center (fig. 10, *B*). The cardiac and vasomotor changes (see fig. 10, *A* and *B*) can in no conceivable way account for the inhibition of the lung motor center which follows the adrenalin injection; for the lungs resume their normal rhythm long before the heart and vasomotor system have escaped from the effects of the adrenalin. We have, furthermore, ruled out the direct or possibly reflex effect of increased amplitude and high blood pressure after adrenalin as being the cause of the inhibition of the lung motor center by injecting adrenalin directly toward the medulla. In such animals the jugular

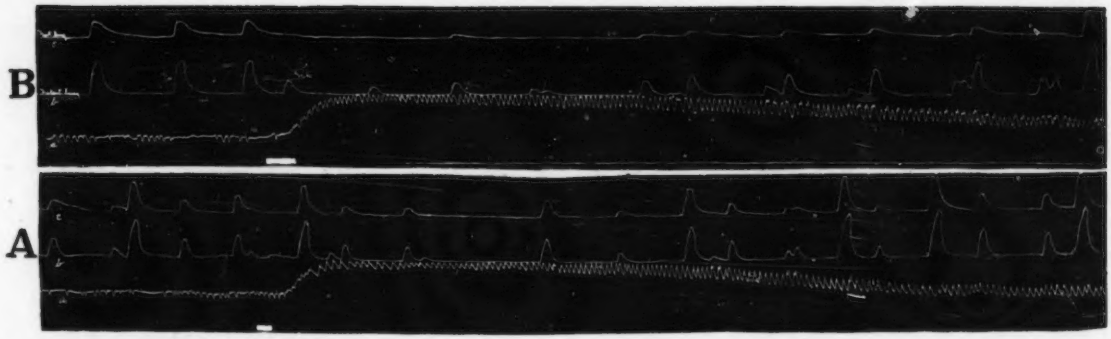

Fig. 10. Water manometer tracings of the intrapulmonic pressure in the turtle. Animals decerebrated and curarized. Left lung isolated except for pulmonary vagi and blood vessels. Cannula in tip of left lung. Record from right lung taken by tracheal cannula; *a*, carotid blood pressure; *b*, tracing from right lung; *c*, tracing from left lung.

A, signal, intravenous injection of 0.1 cc. 1/1000 adrenalin.

B, occlusion of artery and vein to left lung. Signal, intravenous injection of 0.2 cc. 1/1000 adrenalin.

Showing depression of lung motor center (medulla) by adrenalin.

Fig. 11. Water manometer tracings of the intrapulmonic pressure in the turtle. Animal decerebrated and fixed on stand ventral side down. Record from left lung, isolated except for pulmonary vagi and blood vessels. Cannula in tip of lung; left bronchus ligated. Left vagus sectioned in the neck, paralyzing the left lung.

Signal, tetanizing peripheral end of left vagus for 5 seconds at 2-minute intervals.

A, intravenous injections of *a*, 0.5 cc. 1/1000 adrenalin; *b*, 1.0 cc. 1/1000 adrenalin.

B, *x*, intravenous injection of 0.5 cc. 1/1000 adrenalin.

Showing slight peripheral lung depression by very large quantities of adrenalin.

veins and carotids were tied off. The injections of adrenalin were made through a cannula placed in the central end of one carotid, the blood and Ringer's solution escaping from a slit in the external jugular vein on the central side of the ligature. Care was of course exercised in preparing the vagi so that they were not compressed or injured. The injection of adrenalin toward the brain (medulla) effected the same inhibition in the absence of the cardiac and the vascular changes which might affect the medullary center when the drug is injected intravenously.

Since the injection of Ringer's solution or blood-Ringer mixture toward the brain without adrenalin either causes no change in the activity or augments the rhythm and control experiments having shown that the acidity of adrenalin is not responsible for this inhibition, we are forced to the conclusion that inhibition of the lung rhythm following the intravenous injection or medullary perfusion with adrenalin is due directly to the action of the drug.

Small doses of adrenalin, therefore, have the following effect on the respiratory mechanism of the turtle: If the turtle has suffered no loss of blood and is in good physiological condition, adrenalin increases the rate but diminishes the vigor both of external respiratory movements and the contractions of the lungs. In partially asphyxiated preparations or preparations which have deteriorated, adrenalin slows and weakens the lung rhythm. Both augmentation of respiratory rate and lung contractions or slowing of rate of lung contractions and decreased amplitude after adrenalin injection (depending on the physiological condition of the preparation) are brought about essentially by the central action of the drug.

That adrenalin in *large doses* may act peripherally is shown in figure 11, *A* and *B*. In these preparations the injections of 0.5 cc. 1/1000 adrenalin markedly reduced the amplitude of the lung contractions obtained by tetanizing the peripheral end of the vagus every two minutes for five seconds. Even large doses of adrenalin have no effect on the tetanus of the lung brought on by the intravenous injection of large doses of nicotine or histamine. These results are in line with our general conclusion that adrenalin, depending on the condition of the animal, stimulates or depresses the activity of the lung by direct central action and that its peripheral action in small doses is negligible.

In conclusion we point out a fact which the reader has no doubt noted, namely, that the central action of this drug depends on the physiological state of the respiratory and lung motor centers. Under

physiological conditions adrenalin stimulates both the respiratory and the lung motor center. If the centers are partly asphyxiated the same dose of the drug effects a prompt inhibition. These results harmonize with the well-established therapeutic use of this drug in abolishing the bronchial constriction and temporarily relieving the patient from dyspnea in certain cases of bronchial asthma. But there can be no doubt, however, that in certain species of animals adrenalin does exercise more of a peripheral action than it does in the turtle, as shown by Jackson (5) on the dog.

On the basis of the prevailing theory of adrenalin action the slight peripheral depression of the lung motor mechanism by large doses of adrenalin would point to the presence of a sympathetic inhibitory nervous mechanism to the turtle lung. But we do not believe that this conclusion is justified, in absence of more direct proof of the existence of such inhibitory nerves. For it is possible that in large doses adrenalin may have a direct depressor action on the lung motor tissues analogous to that on the respiratory center and the lung motor center when these are below par owing to partial anemia and asphyxia.

II. RESULTS ON THE SNAKE (EUTENIA ELEGANS)

As was noted in our previous communication in the species of snake available for this work the lung musculature is very slightly developed, and mainly confined to the base of the lung (the basal third). Even with the most delicate recording devices the maximum lung contractions appear very feeble. The only drugs investigated on the snake lung were atropin, nicotine and histamine.

Atropin. Atropin in small doses (0.5 to 1.0 mgm.) abolishes the action of the vagi on the lung (fig. 12, *B*, *D*) just as in the turtle. No inhibitory action on the lung from vagus stimulation can be detected after the atropin paralysis. Direct stimulation of the base of the lung after atropin paralysis causes feeble lung contractions, except after very large doses.

Nicotine. Nicotine in relatively small doses (0.5 to 1.0 mgm.) paralyzes the motor action of the vagi. Larger doses of nicotine abolish the lung irritability so that no contractions are produced on direct stimulation with the tetanizing current. The lung motor fibers of the vagi are more promptly paralyzed by nicotine than are the cardiac inhibitory fibers. There is thus a striking difference between the turtle and this species of snake, the efferent lung vagi being readily paralyzed by nicotine in the latter, and practically not at all in the former group.

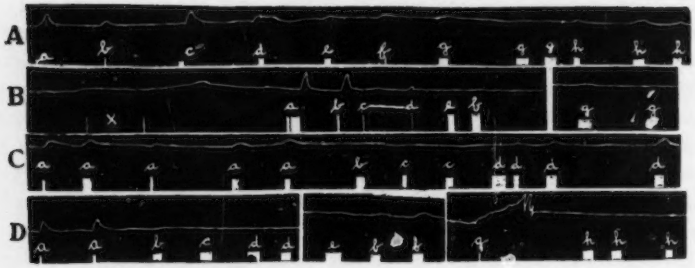

Fig. 12. Tambour tracings of the intrapulmonic pressure in the snake (eutenia elegans). Spinal transection in lower cervical region and pithing of spinal cord. Animal on back. Lung exposed by median ventral incision. Cannula in the thin saclike tip of the single lung. Vagus exposed in neck. All injections through portal vein.

A = *a*, lung contraction following external respiration; *b*, crushed brain with hemostat; *c*, electrical stimulation of peripheral vagus; *d*, intravenous injection ½ mgm. nicotine; *e*, stimulation of peripheral end of vagus; *f*, intravenous injection of ½ mgm. nicotine; *g*, electrical stimulation of peripheral vagus; *h*, electrical stimulation of lung directly with increasingly stronger current.

Showing paralysis of pulmonary motor fibers in the vagus by 1 mgm. nicotine, and contractions of the lung on direct stimulation after nicotinization.

B at *X*, intravenous injection of 0.05 cc. 1/1000 histamine hydrochloride; *a*, electrical stimulation of peripheral end of left vagus; *b*, electrical stimulation of peripheral end of right vagus; *c*, *d*, intravenous injection of 3 mgm. atropin sulfate; *e* and *f*, electrical stimulation of the peripheral end of the left and right vagus respectively; *g*, direct stimulation of the lung.

Showing pulmonary contraction from histamine as well as complete paralysis of the vagus terminations and almost complete paralysis of lung musculature after *large* doses of atropin.

C at *a*, stimulation of peripheral end of the right vagus; *b*, intravenous injection of 1 mgm. atropin; *c*, electrical stimulation of right peripheral vagus with same strength as at *a.*; *d*, direct stimulation of lung musculature showing complete paralysis of vagus motor fibers to the lung by a quantity of atropin insufficient to effect the direct excitability of the pulmonary muscle.

D at *a*, electrical stimulation of the peripheral end of the vagus; *b*, intravenous injection of 2 mgm. nicotine; *c*, electrical stimulation of the peripheral end of the vagus; *d*, electrical stimulation of the peripheral end of the vagus, stronger current; *e*, direct electrical stimulation of the lung; *f*, direct electrical stimulation of the lung, stronger current; *g*, intravenous injection of large dose of atropin (6 mgm.); *h*, direct stimulation of lung with various strengths of currents.

Showing paralyzing action of the motor endings of vagus by a large dose of nicotine with conservation of the direct excitability of muscle. Also the loss of direct excitability of the pulmonary musculature following injection of toxic doses of atropin.

Histamine. Histamine induces a feeble lung hypertonus or contraction, much feebler than the active lung contractions associated with the normal respiratory movements, or that induced by stimulation of the vagi.

It is thus apparent that the action of these drugs on the lung motor mechanism is the same in the turtle and the snake, the noteworthy exception being the ready paralysis by nicotine of the lung motor nerves in the snake, and their great resistance in the turtle.

SUMMARY

1. Curare in small doses depresses temporarily the lung motor mechanism of the turtle by central action. In larger doses it exercises also a slight depressor action on the peripheral structure.

2. Picrotoxin exercises a distinct stimulating action on the lung motor mechanism of the turtle by central action. It accelerates the normal lung rhythm of active preparations and increases the excitability of the center to reflex stimulation. It may induce incomplete tetanus of the lung. None of these effects occurs after vagotomy.

3. Nicotine in small doses causes a central inhibition of the lung motor mechanism of the turtle; in larger doses it produces pronounced lung contraction presumably by direct muscular action. The response of the lung becomes progressively weaker on repeated injections. At a time when nicotine no longer causes lung contraction, stimulation of the peripheral vagus still gives marked motor response. It is not possible to paralyze with nicotine the nerve endings of the motor fibers to the turtle's lung except with doses of nicotine which act as general protoplasmic poison.

4. Histamine causes in the turtle powerful lung contractions by direct peripheral action. It slows slightly the lung rhythm of active preparations by inhibiting the lung motor center. Both actions of histamine become progressively feebler on repeated injections of the drug.

5. Atropine, even in small doses, promptly paralyzes the endings of the motor fibers in the turtle's lung.

6. Adrenalin accelerates the external respiratory movements of the turtle but weakens external respiration and the amplitude of the lung contractions in turtles which are in good condition by acting directly on the respiratory and the lung motor center. In partially asphyxiated animals the same dose of adrenalin promptly depresses the lung motor center. In very large doses adrenalin has a peripheral depressor

effect on the neuro-muscular apparatus of the lung. Such doses have, however, no effect on the lung tetanus induced by large doses of nicotine or histamine. These facts would seem to indicate that the depressor action of this drug on an artificial rhythm induced by tetanizing the peripheral end of the vagus is due to its action on the termination of the vagus nerve; for the same or even large doses of adrenalin have no influence on histamine or nicotine tetanus resulting from the direct muscular stimulation of these drugs.

7. The action of atropin, nicotine and histamine on the lung motor mechanism of the snake is the same as in the turtle as far as we were able to investigate the action of these drugs, with this exception, that nicotine paralyzes the vagus terminations with greater ease than it does in the turtle.

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ADDENDUM: Referring to the action of epinephrin on the lung motor tissues (pp. 26, 27), we have recently found, in connection with our work on the pulmonary vasomotor nerves, that if the turtle lung is in a state of marked spontaneous tonus, epinephrin even in small doses (0.1 cc.–1:1000) induces a primary inhibition of the lung tonus.

January 17, 1921.

STUDIES ON THE VISCERAL SENSORY NERVOUS SYSTEM

V. Cardiac and Vasomotor Reflexes Induced by Visceral Stimulation in Amphibia and Reptilia

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AMPHIBIA (RANA PIPIENS)

Literature. The earliest work on cardiac reflexes in the frog induced by stimulation of visceral afferent nerves appears to be that of Budge. In 1846 he reported that electrical stimulation of the intestines, the lung and the urinary bladder caused inhibition of the heart. But these results were not constant, and mechanical stimulation of these organs, in the hands of Budge (2), had no effect on the heart rate. Nearly twenty years later the problem was re-investigated by Goltz (7), (8), and by Bernstein (1). Goltz showed that gentle mechanical stimulation of the abdominal viscera (tapping the abdomen with the scalpel handle) causes reflex cardiac inhibition. The reaction failed after section of the vagi, or if the animal was anesthetized with chloroform or paralyzed by curare. This became the so-called "Klopf Versuch" of Goltz, the example of inhibitory reflexes performed by students in physiology as a routine teaching experiment. Bernstein showed that the cardiac inhibition described by Goltz was due to the stimulation of the afferent fibers in the mesenteric and abdominal sympathetic nerves, and added the further fact that the reflex takes place through the medulla, the parts of the brain anterior to the medulla not being necessary for the reaction. A year later Goltz (8) reported that mechanical stimulation of the intestines in the frog induces a general vasodilator reflex. Tarchanoff (16) exposed a loop of the frog's intestine to the air by means of a small incision in the abdominal wall. After several hours this exposed loop became hyperemic and when in this condition the slightest touch of the loop with the tip of the finger caused reflex cardiac standstill lasting up to thirty seconds. This slight stimulation of the normal gut did not give the heart reflex. The

heart reflex was abolished by curare and by section of the vagi. Boruttau and Braun (17) showed that the slowing of the heart of the frog on stimulation of the intestines is not a bradycardia due to high blood pressure.

These appear to be the only records in the literature having direct bearing on our present work. There are observations of Pick (14), Nussbaum (12) and Joseph (9) on vasomotor reflexes in the frog induced by stimulation of the skin, and the more recent work of Kuno (10) and of Tawaststjerna (15) on the frog's blood pressure and vasomotor reflexes, but if further work has been done on the problem of visceral sensory action on the cardiac and vasomotor centers in the amphibia we have been unable to locate it.

Experimental methods. The common laboratory frog (Rana pipiens) was used almost exclusively in this work. After decerebration the animal was placed on its back and held in that position by frog clips. The thoracic and abdominal viscera were exposed by an incision in the paramedian line in order to avoid injury to the large median abdominal vein. In some instances a cannula was inserted in the tip of the lung for purposes of rapid inflation and deflation of the lung. When properly prepared, the decerebrated frog will under these conditions not only execute the swallowing movements characteristic of his external respiration but will fill his lungs periodically like a normal intact frog. Our observations on the frog were restricted to the plainly visible changes in the rate of the heart brought about by the stimulation of the various thoracic and abdominal viscera. The results were so pronounced and decisive that no attempt was made to record them graphically.

Results: I. Frog (Rana pipiens). Rapid inflation or deflation of the lungs did not have the slightest effect on the rate of the heart. This seemed quite strange for electrical or mechanical stimulation of the tip of the lung effected promptly a more or less prolonged cardiac inhibition. The mechanical (touching, rubbing, pinching, crushing) or electrical stimulation of all other viscera, with exception of the oviducts and ovaries themselves, causes prompt cardiac standstill of variable duration. Especially powerful in this respect was mechanical or electrical stimulation of the bladder, spleen and small intestine although electrical stimulation of the stomach and large intestine effected a more or less prompt cardiac inhibition. Cardiac standstill was not readily obtained from mechanical or electrical stimulation of the peritoneum, gall bladder region and kidney. The complete and

almost instantaneous inhibition of the heart following the mere touching of the urinary bladder and small intestine was particularly striking. In striking contrast with these results was the ineffectiveness of stimulation of the cornea and anterior nares. Electrical stimulation of the mesentery likewise gave powerful reflex cardiac inhibition.

II. Cryptobranchus. A few observations were made on this form of gill-less salamander. These observations were made incidental to a study of lung reflexes in this amphibian. The results were similar to those obtained in the frog. Electrical stimulation of the skin, toes, urinary bladder, small intestine, large intestine and rectum gave pronounced cardiac inhibition. Even stimulation of the mesentery of the ovaries which in the turtle and frog yielded negative results caused in this animal striking cardiac standstill. In this animal also the stimulation of the anterior nares had no effect on the rate of the heart.

In no instance did we observe reflex cardiac acceleration from the stimulation of the visceral nerves in either the frog or in cryptobranchus. These results confirm and extend the old observations of Budge, Goltz and Bernstein.

The observation of Goltz that the cardio-inhibitory reflex fails in curarized frogs is also confirmed. The vagus cardio-inhibitory mechanism is promptly paralyzed by curare long before the paralysis of the skeletal neuro-muscular mechanism and even by doses of curare too small to cause complete paralysis of the latter.

The curare paralysis of the cardiac vagus leaves the cardio-accelerator fibers apparently intact, because stimulation of the peripheral end of the vago-sympathetic nerve produces the acceleration of the pulse typical after atropin or nicotine paralysis of the inhibitory mechanism.

Nevertheless, we have not obtained in the curarized frog definite reflex cardiac acceleration from stimuli that cause reflex cardiac inhibition in the non-curarized animal. Curare must, therefore, induce depression at some point in the accelerator reflex path, or else we must conclude that in the frog the cardio-accelerator reflex center is less readily accessible from the visceral afferent nerves.

REPTILIA

Experimental methods. All animals were decerebrated, and no anesthetics were employed. Most of the work was made with animals prepared and fixed in the turtle stand in their normal position (ventral

side down), as described by us in a previous report (3), the blood pressure and pulse records being taken from the carotid artery (mercury manometer). This involves the least dissection and consequent interference with normal reflexes from sensory traumatic stimuli. In some cases the blood pressure and pulse were recorded from one of the innominate arteries, or from the main artery of the hind leg.

In the early part of the work several experiments were made on decerebrated turtles fixed dorsal side down. After removing the plastron, the cardiac reflexes on visceral stimulation were recorded by direct observation.

The size of the animals used in this work required a size of carotid blood pressure cannula of less than a millimeter bore. We used a straight glass cannula filled with a 5 per cent solution of sodium citrate. But with a cannula of this small bore there was considerable difficulty in the way of blood coagulation in the neck of the cannula, especially in animals with low blood pressure.

The main difficulties are low blood pressure due to loss of blood in removing the dorsal shell sufficiently for direct stimulation of the various visceral organs; and the tendency of the turtle's arteries to go into prolonged local contracture on direct mechanical handling. These contractures may last from 15 to 60 minutes or longer. It is therefore essential to adjust the blood pressure recording with the least possible direct handling of the artery used, both in inserting the cannula and in freeing the artery from adjacent tissues. Low blood pressure can be temporarily overcome by intravenous injection of Ringer's solution with small amounts of adrenalin. The blood pressure raising action of this drug in the turtle may last for 30 to 60 minutes. When all unfavorable factors are reduced to the minimum, one can secure a continuous blood pressure and pulse record from the turtle's carotid for 5 to 15 hours without coagulation interference. But in prolonged experiments, with much of the viscera exposed and traumatized, the great lowering of the blood pressure due to transudation of the blood plasma into the tissue and lymph spaces is not easily controlled.

The carotid blood pressure in these turtles varied from 30 to 90 mm. Hg. at the start of an experiment. This is considerably higher than that reported by Kuno (10) for the frog (30 mm. Hg.). The highest pressures (no Ringer's solution or blood pressure raising drugs) approach the lower normal blood pressure level in birds and mammals. If the heart rate is slow the pulse pressure may be as high as 15 to 20 mm. Hg. Ordinarily it is less than 10 mm. Hg. (4 to 8 mm. Hg.). With a

very rapid heart the pulse pressure may be too slight to be recorded with the mercury manometer.

In preparations with blood pressures down to 5 or 10 mm. Hg. we found it expedient to attach a delicate tambour to the arm of the manometer which ordinarily contained the float with writing point. The delicate tambour with its compound lever will record the rate and strength of the heart beat, but not the blood pressure, with satisfactory accuracy.

Cardiac arrhythmia. If the decerebrated turtle is prepared with the minimum trauma and loss of blood the pulse is usually very regular except on experimental interference. On the other hand, low blood pressure, contracture of the arteries or imperfect lung ventilation are usually accompanied by various types of cardiac arrhythmias that may persist for hours. We shall later show that these persistent arrhythmias are essentially of vagal origin, and due to partial asphyxia. The arrhythmias consist of two or three beats followed by a vagal pause in regular sequence. A slow but perfectly regular vagus pulse is less frequently seen in animals prepared according to our method.

These "regular irregularities" of the turtle heart in partial asphyxia or other types of continuous medullary stimulation seem to indicate a kind of "see-saw" action between the cardio-inhibitory and the cardio-accelerator centers or else a regular rhythm in the vagus action on the heart.

RESULTS

I. Cardio-vascular reflexes from the respiratory tract:

a: Influence of the spontaneous respiratory movements. The spontaneous respiratory movements may have no effect on the heart rate or the blood pressure (fig. 1, *A*). This is true particularly if the respiratory movements are very slight or feeble, from ample aeration of the blood. When the respiratory movements are more powerful the heart is always affected either in the direction of acceleration or inhibition (fig. 1, *B*, *C*, *D*.) We are speaking now of the external respiratory movements as distinct from the contraction of the lung musculature that normally follows these movements.

These cardiac effects may be due, *a*, to a spread of the respiratory nervous impulses from the respiratory to the cardio-regulative centers in the medulla; *b*, reflexes from sensory nerves in the lungs; *c*, reflexes from sensory nerves in the respiratory muscles. We cannot exclude any of these factors completely. In the intact animal all three may be

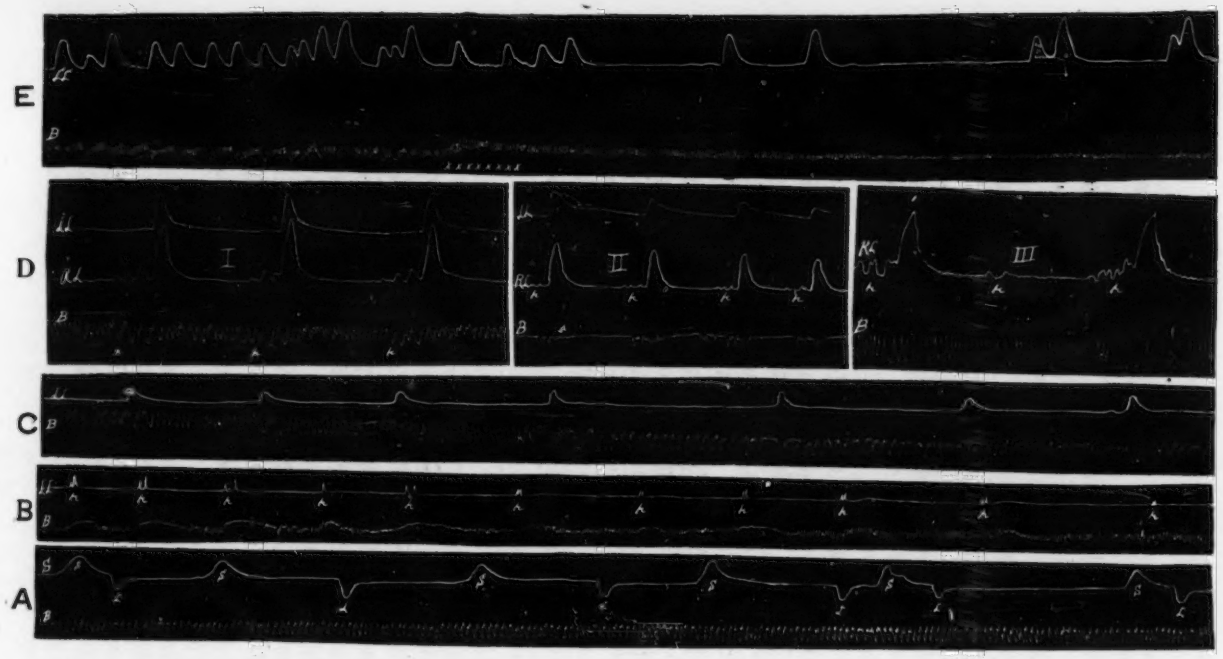

Fig. 1. Records of carotid blood pressure, respiration and lung contractions (water manometer) in decerebrated turtles.

B, carotid blood pressure; *L.L.*, contractions from left lung isolated except for pulmonary vagi and blood vessels. Cannula in tip of lung, left bronchus ligated. *L.R.*, record from right lung (tracheal cannula) intact; *S*, record from empty stomach (balloon method); *r*, external respiratory movements.

A, shell intact, persistent cardiac arrhythmia; *s*, stomach contractions; *L*, lung contractions preceded by respiratory movements too feeble to be recorded on stomach tracing. Showing no influence of respiratory and lung rhythm on heart.

B, spontaneous respirations but no lung contractions. Showing cardiac acceleration and rise in blood pressure with each respiratory group.

C, Animal partially curarized. Showing both cardiac acceleration and inhibition synchronous with respiration and lung contractions.

D, Animal partially curarized. Showing cardiac inhibition during respiration and lung contractions.

E, Animal completely curarized and incomplete asphyxia; *x–x*, artificial respiration in intact lung. Showing cardiac inhibition during respiratory center discharge and lung contraction in partial asphyxia, and absence of cardiac effects after eliminating asphyxia by a period of artificial respiration.

involved. But the fact that the same results on the heart may be obtained in a completely curarized animal, goes to show that the spread of the nervous discharges from the respiratory to the cardiac centers of the medulla is a real factor. In a curarized animal we can follow the rhythmic activity of the respiratory center by the lung contractions that appear as side events in the external respiratory act (3).

It is difficult to determine whether the discharge of the lung motor center also may spread to the cardiac centers. The cardiac effects from the discharge of the respiratory center may outlast the external respiratory acts and thus appear, in part, parallel with the contraction of the lungs (fig. 1, *D*) without necessarily being causally connected with that act or mechanism. But in many cases the respiratory effect

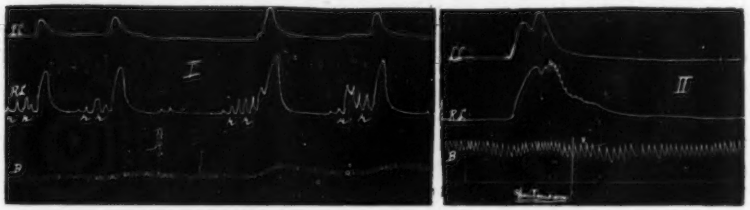

Fig. 2. Records of carotid blood pressure and lung contractions (water manometer) in the turtle.

L.L., left lung isolated except for pulmonary vagi and blood vessels; *R.L.*, right lung (intact); *B*, carotid blood pressure; *r*, respiratory movements.

I, Animal partially curarized. Showing rise in blood pressure (vasomotor action) accompanying respiratory and lung motor actions.

II, Animal completely curarized; no skeletal muscle movement present. Showing cardiac inhibition preceding and at the end of the spontaneous lung contractions.

on the heart is so transitory that it passes before the lung contractions actually appear, and in such cases there is rarely any cardiac irregularity associated with or resulting from the lung contractions.

There appears to be an exception to this rule, in the case of marked asphyxia. In figure 1, *E* (animal completely curarized and in marked asphyxia) there are definite cardiac effects parallel with the lung contractions. But the lung contractions follow in so rapid succession that there is obviously a very rapid and powerful discharge from the respiratory center. After removal of the asphyxia by artificial respiration the lung contractions fail to influence the heart, as shown by the end of this record.

When the respiratory act causes cardiac acceleration there is usually some rise in blood pressure that may be caused by the faster heart beat. Less frequently do we find rise in blood pressure without heart change parallel with the respiration and lung contractions (fig. 2, *I*).

It is thus clear that in the decerebrated turtle the discharge from the respiratory center may spread into the cardiac centers causing either cardiac inhibition or acceleration, temporarily. The stronger the respiratory discharge or the greater the excitability of the cardiac centers, as in asphyxia, the greater the disturbance of the heart by the respiratory discharge. Ordinarily the lung contractions following the respiratory act have no effect on the heart, but if the cardiac centers are hyper-irritable, as in asphyxia, these lung contractions may affect the heart, either by sensory impulses from the lungs or as a result of spread of the nervous discharge from the lung motor to the cardiac centers. The first possibility might be eliminated by mild cocainization of the pulmonary vagi.

b. Cardiac reflexes from inflation and deflation of the lungs. Inflation and deflation of the lungs in artificial respiration causes acceleration of the heart (fig. 1, *E;* fig. 3, *A*, *B*, *C*). There are probably two factors involved in this reaction. If the heart is slow or irregular from excessive vagus tonus caused by partial asphyxia, the aeration of the blood by the artificial respiration will itself accelerate the heart by diminishing the vagus tonus. In such preparations section of both vagi nerves causes the same type of cardiac acceleration permanently, as is induced by artificial respiration temporarily (fig. 3, *E*, *I*.). But still deeper asphyxia causes cardiac irregularity by a direct action on the peripheral heart motor mechanism, and this irregularity (slowing) can be removed by artificial respiration in the denervated lungs, thus excluding afferent impulses from the lungs and vagi action on the heart (fig. 3, *E*, *II*).

It can readily be shown, however, that inflation or deflation of the lungs affects the heart reflexly, independent of its effect on CO_2 and O_2 content of the blood. A single inflation of one lung with air, carbon dioxide or hydrogen causes a temporary acceleration of the heart (fig. 4, *A*, *B*, *C*). Moreover, a single inflation of one lung with air, after previous occlusion of the blood vessels of this lung produces cardiac acceleration (fig. 4, *D*, *II*). Primary inhibition of the heart induced reflexly by inflation of the lungs was never obtained, the lung inflation always accelerating the heart rhythm, probably by decreasing the vagus tonus and stimulating the cardio-accelerator

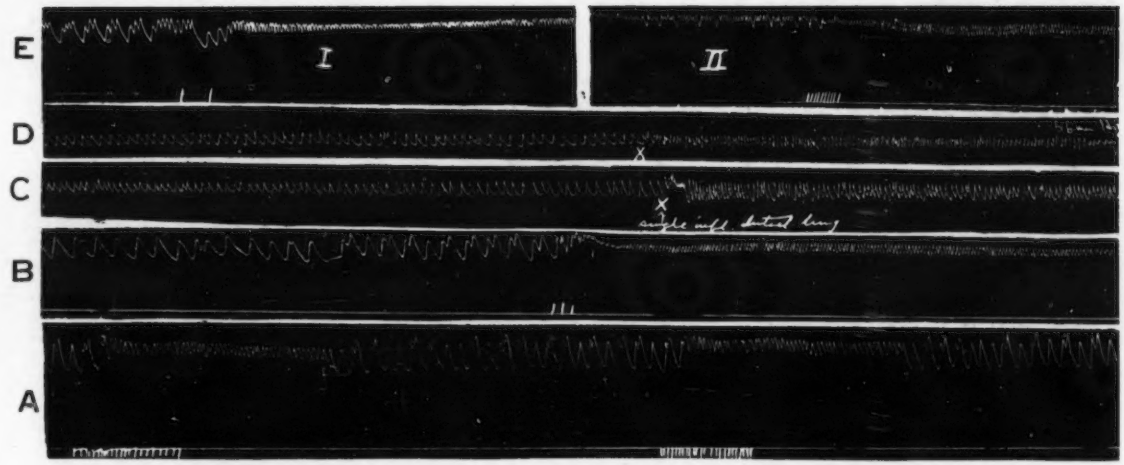

Fig. 3. Records of carotid blood pressure in the decerebrated turtle. Vagi intact except as indicated in *E*. Vagus heart, slow and regular or slow and irregular, due to partial asphyxia, due to absence of or interference with external respiration. *A*, *B* and *C*, animals partially curarized; *E*, animal completely curarized; *D*, no curare, absence of respiratory movements probably due to "shock" from removing shell on left side.

A, signal, groups of artificial respirations; *B*, signal, three inflations of one lung; *C–x*, single inflation of one lung; *D–x*, single spontaneous respiration; *E–I*, signal section of both vagi in neck; *E–II*, both vagi sectioned in the neck. Signal, artificial respiration. Showing removal of central cause of cardiac asphyxial arrhythmia by spontaneous or artificial respiration; and by vagi section, and peripheral asphyxia arrhythmia removed by artificial respiration through the denervated lungs.

mechanism. Strong deflation of the lungs, on the other hand, tends to produce a temporary inhibition of the heart (fig. 4, *E*). This reaction is not produced if the isolated (except pulmonary vagi and blood vessels) and inflated lung is allowed to collapse until it is completely empty of air. It is produced by suction through the trachea or bronchus of an intact lung. It is therefore uncertain whether the reflex heart inhibition following strong deflation of an intact lung is due solely to the stimulation of sensory fibers in the lung itself, as on account of the mode of fixation of the lung in the body cavity such suction will cause lowering of the intra-abdominal pressure and greatly altered tension, especially on the organs connected with the membranes that support the lung. This question could be decided by isolating one lung, so that it would not be passively influenced by deflation of the other lung, then deflation of this lung after section of the vagus on that side, leaving the vagus on the side of the intact lung as the efferent pathway to heart reflexes that the deflation might induce by stimulation of visceral sensory nerves other than those in the lung itself.

c. Cardiac reflexes from direct stimulation of the pulmonary vagi. Direct stimulation of the central end of the pulmonary vagi induced heart acceleration together with a tendency to fall of blood pressure (fig. 5, *A*). At no time did we obtain reflex heart inhibition from this stimulation. As a rule the heart acceleration persists for a considerable period after cessation of the vagus stimulation.

When the cardiac vagi are cut on both sides stimulation of the central end of the pulmonary vagi usually causes a temporary rise in the blood pressure, followed by a more marked fall, even during the stimulation (fig. 5, *C*). Less frequently the stimulation may induce a rise in blood pressure only. Evidently the pulmonary afferents have both pressor and depressor actions on the vasomotor mechanism.

The nervous relations between the cardio-vascular and respiratory mechanisms are, therefore, very complex. There may be a spread of the discharge from the respiratory center to the cardiac centers causing either heart inhibition or heart acceleration. There may be cardiac and vasomotor changes associated with the lung contractions, either through spread of the centrifugal impulse from the lung motor center, or through afferent impulses via the vagi induced by the lung contractions. Lung inflation (artificial or spontaneous respiration) may stimulate afferent fibers primarily the pulmonary, leading to cardiac acceleration, by reducing the tonus of the cardiac vagi. Lung deflation causes cardiac inhibition, probably in part by mechanical stimulation

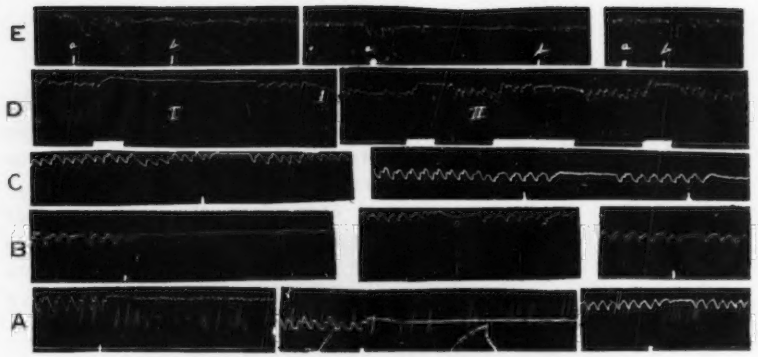

Fig. 4. Records of carotid blood pressure in decerebrated turtles. *A–D*, animal fixed on dorsal side, left lung isolated except for pulmonary vagi and blood vessels. All inflations in isolated lung. *E*, animal fixed dorsal side down and plastron removed; suction on lung through trachea.

A, signal, single inflation of lung with air; *B*, signal, single inflation of lung with CO_2; *C*, signal, single inflation of lung with H_2; *D*, signal, single inflation (holding pressure) during period indicated. *I*, lung blood vessels free; *II*, same preparation after occlusion of lung blood vessels; *E*, suction on lung through trachea from *a* to *b*. Showing presence of sensory fibers in the lung whose stimulation by distention causes cardiac acceleration, and by collapse of the lungs, and traction on supporting structures, cardiac inhibition.

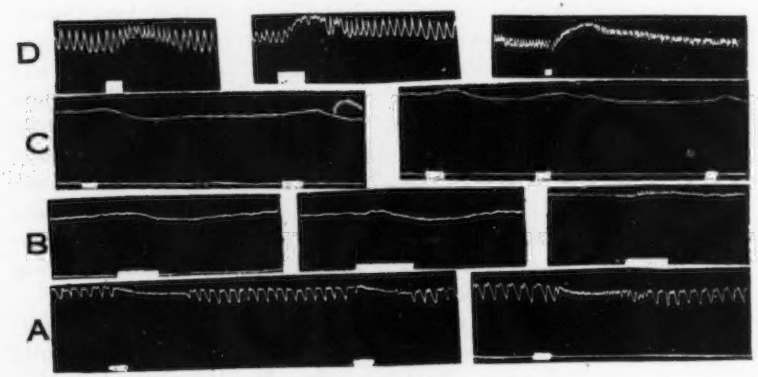

Fig. 5. Records of carotid blood pressure in decerebrated turtles, light curare.

A, cardiac vagi intact. Signal, tetanization of central end of right pulmonary vagus. Showing cardiac acceleration and tendency to fall of blood pressure.

B, cardiac branches of both vagi severed. Signal, tetanization of central end of right pulmonary vagus. Showing fall and rise in blood pressure but no effect on heart.

C, both vagi severed in neck. Signal, tetanization of central end of left vagus. Showing primary vasoconstriction, with subsequent tendency to vasodilatation. No effect on heart.

D, Vagi intact. Mechanical stimulation of the posterior nares. Showing cardiac acceleration and vasoconstriction.

of the afferent nerves outside the respiratory tract. And, lastly, direct stimulation of the central end of the pulmonary vagi causes reflex heart acceleration and pressor and depressor effects on the vascular mechanism, apart from the heart reflex.

Apart from the pulmonary vagi themselves, the cardio-vascular action of the afferent nerves to other parts of the respiratory tract was not studied, except for the posterior nares. Mechanical stimulation of the posterior nares invariably caused acceleration of the heart and a rise in the blood pressure (fig. 5, *D*).

II. Cardio vascular reflexes from the alimentary tract:

a. Cardio-vascular reflexes from the stomach. Several years ago it was noted by one of us in man that the strong contractions (hunger contractions) of the empty stomach were accompanied by cardiac and vasomotor disturbances (5). It was assumed that these were induced by impulses from the gastric afferent nerves, the stimulus to these nerves being the stomach contractions. There is other evidence in the literature that the gastric afferent nerves, especially when stimulated by tension or contraction, may act reflexly on the cardio-vascular mechanism in the mammal. Hence we made a very extensive study in the decerebrated turtle of the possible reflex influence of the normal contractions of the empty stomach on the heart and the blood vessels.

In all of the experiments in this group the animals were decerebrated and fixed on the turtle stand in the usual way. The shell was usually left intact, as it was found that the greater the bodily trauma to the animal the smaller the chances of obtaining the normal vigorous and regular contractions of the empty stomach, described by Patterson (13). The stomach balloon was introduced through a slit in the esophagus and the cannula fixed in the carotid artery, with minimum dissection and trauma, especial care being taken not to touch or otherwise injure the vagi nerves. In a few experiments the dorsal shell was removed on one side for the purpose of isolation of one lung.

In our best preparations we secured records of the regular and vigorous gastric hunger contractions parallel with the records of carotid blood pressure and pulse, the preparations breathing spontaneously and satisfactorily by means of the two lungs or the one intact lung.

The results of these experiments were essentially negative, the spontaneous contractions of the empty stomach in the decerebrated turtle having little or no reflex influence on the heart or on the blood pressure. Typical tracings are reproduced in figure 6. In some cases there is a slight acceleration of the heart, evidently due to inhibition

of the vagus inhibitory tonus, associated with the contractions (either the contraction phase or the relaxation phase) (fig. 6, *C*), but in other cases of equally strong gastric contractions no definite reflex influence on the heart or the blood vessels appears on the record.

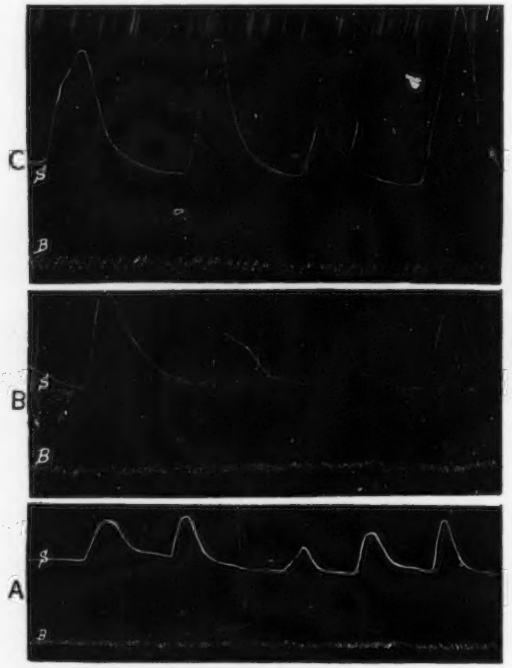

Fig. 6. Simultaneous records of carotid blood pressure and contractions of the empty stomach in decerebrated turtles.

B, carotid blood pressure; *S*, stomach contractions (balloon method) Showing no reflex effects of the spontaneous contractions of the empty stomach on the heart and the blood pressure.

Since there are in the turtle gastric afferent fibers that can influence the heart and the vasomotor mechanism reflexly, our essentially negative results from this type of experiments must mean either that these nerves are not adequately stimulated by the spontaneous stomach contractions, or that the decerebration interferes with conduction cen-

trally by depression of lower centers or actual severance of reflex paths.

Strong distention of the stomach wall by balloon inflation and direct stimulation of the central end of the gastric vagi causes cardiac acceleration and rise in blood pressure (fig. 7). The cardiac reflex is not obtained after section of the cardiac vagi, but the reflex rise in blood pressure can still be produced, showing that the gastric afferent nerves connect both with the cardiac and the vasomotor centers. The cardiac acceleration induced by tetanization of the central end of the gastric vagi appears to be due mainly to depression of the inhibitory tonus of the cardiac vagus.

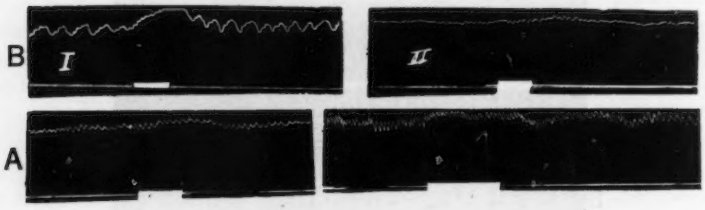

Fig. 7. Records of carotid blood pressure in the decerebrated turtle.

A, signal, distention of empty stomach by inflation of balloon in stomach. Showing slight acceleration of the heart.

B, Animal curarized; *I*, tetanization of central end of the right gastric vagus. Showing heart accelerations and rise of blood pressure; *II*, all vagi branches to heart sectioned. Signal, tetanization of central end of right gastric vagus. Showing rise in blood pressure but no action on heart.

b. Cardiac and vasomotor reflexes from the small intestines. Stimulation of the small intestines causes reflex inhibition or reflex acceleration of the heart (fig. 8). These reactions are obtained both with mechanical and electrical stimulation of the intestinal mesentery. The reflexes are readily induced by the gentlest mechanical stimulation of the surface of the intestine, provided always that the animal is not in shock or otherwise depressed. As the preparation deteriorates the cardiac reflexes from the gut disappear long before the abolition of the skeletal reflexes.

One cannot predict whether a given type or strength of intestinal stimulation will cause an inhibition or an acceleration of the heart, with parallel changes in the blood pressure. In general the cardio-inhibitory reflexes are the predominant ones in non-curarized, and the

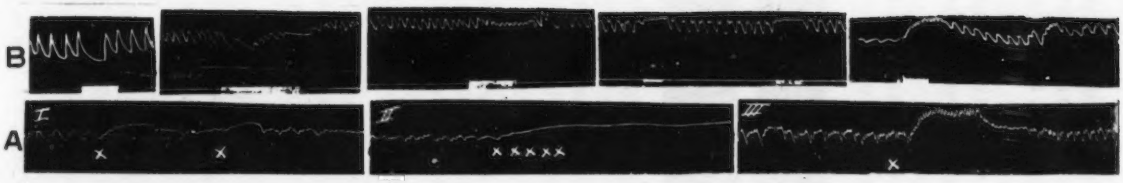

Fig. 8. Records of carotid blood pressure in completely curarized turtles.

A–I, *x*, picking up a loop of small intestine between the fingers; *II*, *x*, stroking a loop of small intestines with the finger; *III*, *x*, crushing a loop of small intestines with forceps. Showing reflex cardiac acceleration and rise in blood pressure.

B, Signal, stimulation of loops of small intestine with the tetanizing current. Showing reflex cardiac inhibition and acceleration.

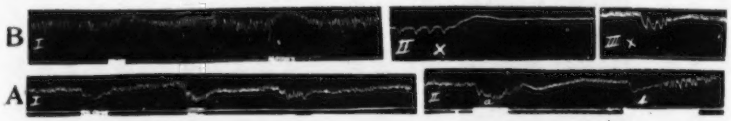

Fig. 9. Records of the carotid blood pressure in the decerebrated turtle.

A, No curare. *I*, signal, distention of rectum and large intestine by balloon; *II*, *a*, pinching large intestine with forceps; *b*, pinching rectum with forceps. Showing reflex vasodilatation and tendency to cardiac inhibition.

B–I and *II*, Animals curarized; *I*, signal, tetanization of the large intestine. *II*, *x*, crushing rectum with artery clamp; *III*, no curare; *x*, mechanical stimulation of the anus. Showing reflex cardiac inhibition and acceleration.

accelerator reflexes in the curarized preparations. But both types of reflexes were obtained in curarized and in non-curarized animals. It may be a question of relative tonus or fatigue of the two cardiac centers.

Distention of the gut by inflation as a type of intestinal stimulation was not tried. Nor did we study the pure vasomotor reflexes from the small intestines after section of the cardiac vagi.

c. Cardiac and vasomotor reflexes from the large intestines and rectum. Mechanical (distention, stroking, cutting, pinching, crushing) or elec-

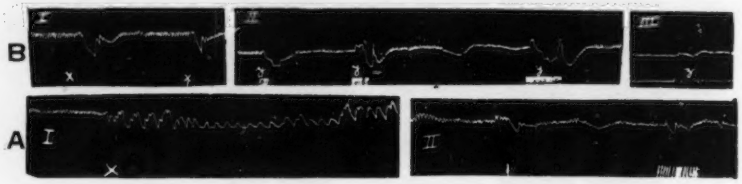

Fig. 10. Tracings of the general arterial blood pressure of the turtle. Turtle decerebrated.

A, I, turtle ventral side down on holder; blood pressure from carotid; dorsal window; *A, II*, animal on dorsal side, plastron removed, blood pressure from innominate.

A, I, at *X*, touching the urinary bladder.

A, II, Signal magnet records mechanical stretching of urinary bladder.

B, I, turtle ventral side down on holder, blood pressure from carotid, dorsal window; *B, II*, and *B, III*, animal on dorsal side, plastron removed, blood pressure from innominate.

x, Cut slit into urinary bladder; *y*, distention of rubber balloon in urinary bladder; *z*, stretching of urinary bladder. Showing the usual cardiac inhibition with fall of blood pressure, which follows mechanical stimulation of the urinary bladder except at *z* a small rise in blood pressure followed stretching of the walls of this viscus.

trical stimulation of the large intestine and rectum causes reflex inhibition or acceleration of the heart (fig. 9). Usually parallel with these reflexes there is a fall or a rise in the blood pressure, but the vasomotor reflexes are to a certain extent independent of the cardiac, as one may obtain a fall of blood pressure with little or no change in the heart rhythm. As in the case of the small intestine, the cardio-inhibitory reflexes predominate in the non-curarized, and the cardio-augmentor in the curarized preparations.

It is thus evident that the stomach and the entire length of the intestines (including the rectum) are provided with afferent nerves,

connecting reflexly with the heart and the vasomotor centers. In the case of the intestines their stimulation gives both inhibitory (and depressor) and accelerator (and pressor) effects. The spontaneous contractions of the empty stomach usually fail to call into play these reflex mechanisms, at least in the decerebrated animal.

III. Cardio-vascular reflexes from the genito-urinary tract:

a. Urinary bladder. Of all the viscera in the abdominal cavity the urinary bladder yields with greatest certainty on stimulation marked reflex effects on the heart and vasomotor system. In any animal in good condition and suitably prepared the mere mechanical touching of this organ may cause promptly a long lasting cardiac inhibition and arhythmia with lowering of the general blood pressure as is shown in figure 10 *A*, *I*. Such an irregularity and slowing of the heart may under certain conditions be even more pronounced and long lasting, than the inhibition effected temporarily by mechanically stretching the horn of the bladder (fig. 10, *A*, *II*). The more violent mechanical stimulation of cutting a slit into the bladder may cause promptly complete cardiac standstill with drop in the general arterial pressure (fig. 10, *B*, *I*, at *x*). Mechanical distention of a rubber balloon in the urinary bladder gives the same result (fig. 10, *B*, *II*, at *y*).

It would appear that mechanical stimulation of any kind gives invariably reflex cardiac inhibition with drop in blood pressure. Occasionally, however, mechanical stimulation (such as stretching) effects a slight rise in general blood pressure (fig. 10, *B*, *III*, at *z*).

The results following electrical stimulation of the horn or neck of the urinary bladder appear to depend on whether or not the turtle is curarized. In the *non-curarized* turtle electrical stimulation of the bladder with a tetanizing current initiates a more or less prolonged cardiac irregularity of vagal origin with some lowering of the general arterial pressure (fig. 11, *A* at *a*); or the cardiac inhibition may become more or less complete during the period of tetanization with no marked tendency to return promptly to the rapid rate characteristic of the heart before the stimulation was commenced. It is of some importance to note that the arterial pressure rises following the stimulation, not only to its original level but may exceed the latter in spite of the persistence of vagus beat (fig. 11, *A* at *b*). These facts might lead us to suppose that the bladder contains pressor and depressor fibers besides those fibers which throw into activity the cardio-inhibitory mechanism. Of these fibers the depressor and those which affect reflex cardiac inhibition predominate in the action on the medullary centers when the

urinary bladder of a non-curarized turtle is tetanized with a strong electrical current. Before the cardiac inhibition has completely disappeared the effect of stimulation of pressor fibers now comes to light, the result being a rise in the mean arterial blood pressure above the original level. If the cardiac inhibitory mechanism is rendered less

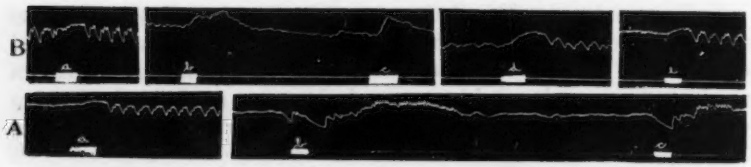

Fig. 11. Tracings of the general arterial blood pressure of the turtle. Turtle decerebrated, on dorsal side, plastron removed, blood pressure from the innominate artery.

A, Tracings taken from *non-curarized* animals. *a*, electrical stimulation of the horn of the urinary bladder with a tetanizing current; *b* and *c*, stimulation of the neck and horns of the urinary bladder respectively with a strong tetanizing current. Showing marked cardiac inhibition and irregularity with lowering of blood pressure as a result of electrical stimulation of the afferent nerves of the urinary bladder.

B, tracings taken from partially *curarized* animals. *a–e*, Electrical stimulations of the neck of the bladder with a strong tetanizing current. Showing marked cardiac acceleration and increase in blood pressure following stimulation of the afferent nerves of the urinary bladder with a strong tetanizing current.

Fig. 12. Tracings of the general arterial blood pressure of the turtle. Turtle decerebrated, blood pressure from carotid or innominate showing at *a* and *b*, cardiac inhibition and drop in blood pressure on mechanical stimulation of the cloaca in the non-curarized animal; at *c*, the reverse effects on mechanical stimulation of the cloaca in the curarized animal.

responsive to reflex stimulation by curarizing the animal, the immediate effect of stimulating the urinary bladder is a pronounced cardiac acceleration with or without a sharp rise in the mean arterial pressure as is seen in figure 11, *B*, at *a*, *b*, *c* and *d*. In such animals the initial cardiac acceleration may be followed by a cardiac inhibition and irregularity associated with a tendency toward a diminution in the general

arterial pressure. It would seem, therefore, that curare reverses the order of appearance of the cardio-vascular responses following electrical stimulation of the bladder or suspends entirely the usual and predominating cardiac inhibition by its powerful depressing action on the cardio-inhibitory mechanism of the heart.

b. Cloaca. The results on the heart and vascular system following mechanical or electrical stimulation of the cloaca are virtually identical with those obtained from the bladder. Here, too, stimulation of the cloaca in the *non-curarized* turtle causes a marked cardiac inhibition with drop in blood pressure. Such stimulation may or may not induce a more or less permanent cardiac irregularity (fig. 12, at *a* and *b*). The blood pressure may and commonly does exceed its original level. If the animal is curarized the same form of stimulation leads promptly to a marked cardiac acceleration accompanied by an appreciable rise in the blood pressure.

c. Kidney, ureter and testis. Mechanical and electrical stimulation of these organs yielded in the few experiments which we tried uncertain results. In general, a rise in pressure with cardiac acceleration followed mechanical or electrical stimulation of these organs.

IV. Cardiac and vascular reflexes from direct stimulation of the central and of the main sympathetic nerve trunks: Our work in this field was much less extensive than that on the visceral organs themselves. Tetanization of the central end of the cervical sympathetic nerve nearly always causes a rise in blood pressure, with or without acceleration of the heart (fig. 13, *A*). The same type of stimulation of the central end of the main abdominal sympathetic nerve trunks causes rise in blood pressure with cardiac acceleration, rarely an inhibition of the heart with a fall in blood pressure. We have seen that the depressor reflexes are ordinarily the ones most readily evoked by the stimulation of the visceral organs themselves (excepting the lungs and the stomach).

In figure 14 a few typical blood pressure records are reproduced from curarized and non-curarized preparations showing the similar or practical identity of the cardio-vascular reflexes evoked from the cutaneous spinal and from the visceral sensory nerves.

Figure 13, *B*, illustrates a source of error that may complicate this line of investigation in the turtle in case the decerebration is incomplete. The turtle was decerebrated, presumably completely, 24 hours before the beginning of the experiments. Cardiac inhibitions appeared that could not be accounted for, and it was found that they were due to visual stimuli, persons moving about in front of the turtle, and

especially by suddenly moving the hand or any object toward the turtle's head. The animal was not curarized. It was breathing spontaneously and regularly. Movements of objects toward the head did not cause movements of the legs of the turtle, although they induced marked cardiac inhibition.

It was found that the posterior end of the cerebral hemispheres had not been completely destroyed. This visual cardiac reflex was never observed in completely decerebrated turtles.

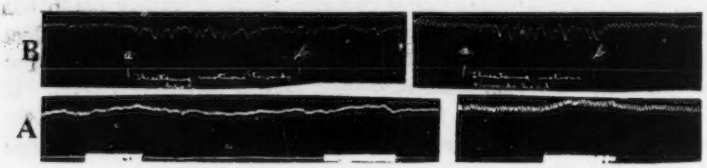

Fig. 13. Records of the carotid blood pressure in the decerebrated turtle.

A, animal curarized. Signal, tetanization of the central end of the cervical sympathetic nerve. Showing reflex rise in blood pressure.

B, No curare; 24 hours after incomplete decerebration; *a* to *b*, making threatening movements toward the turtle's head. Showing reflex cardiac inhibition from visual stimulation.

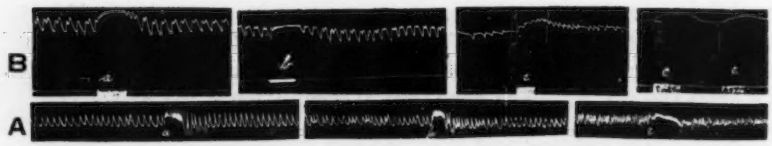

Fig. 14. Records of carotid blood pressure in the decerebrated turtle.

A, No curare; *a*, stroking skin of leg; *b*, pinching toes; *c*, cutting skin.

B, Complete curarization; *a*, tetanization of central end of sciatic nerve; *b*, tetanization of a loop of the small intestine; *c*, tetanization of central end of sciatic nerve after section of both vagi in neck. Showing reflex cardiac acceleration and rise in blood pressure.

V. The reversal of cardio-vascular reflexes after curare: As early as 1856, Kölliker (18) reported that curare paralyzes the inhibitory action of the vagi on the frog's heart as well as the spinal nerves acting on the lymph heart. In larger doses even the vasomotor nerves in the sciatic are paralyzed. According to the investigations of Dale and Laidlaw (6) and Langley (11), curare apparently paralyzes, in the mammal, the sympathetic ganglion cells, particularly those of the heart,

concerned with the transmission of the inhibitory impulses of the vagus to the cardiac musculature. If this drug acts similarly in the turtle we can readily understand why cardiac inhibition or arrhythmia with a drop in blood pressure which so commonly follows stimulation of afferent visceral fibers in the non-curarized turtle, is absent in the curarized preparation. In the non-curarized animal stimulation of visceral afferent fibers causes such profound cardiac inhibition and drop in blood pressure that the latter overpowers the cardiac accelerators and obscures the vasoconstriction, which may appear as a slight after effect. But when the peripheral ganglia along the course of the vagus are depressed by curare the cardio-accelerator and vasoconstrictor effects become predominant, for the inhibitory impulses no longer reach the heart via the vagi. As a result the immediate effect of the stimulation of visceral afferent fibers in the *curarized* animal is a marked rise in the general arterial pressure with cardiac acceleration.

The fact that curare in the turtle primarily stimulates the cardiac vagus mechanism followed by a more or less permanent cardiac acceleration (4)[1] renders it probable that this drug acts the same in the turtle as in the mammal.

SUMMARY

1. In the frog (Rana pipiens) stimulation of the lungs, the gastro-intestinal and the genito-urinary tracts causes reflex cardiac inhibition. No cardio-accelerator action is induced by these stimulations. In curarized preparations the reflex fails owing to vagus paralysis.

2. In the salamander (cryptobranchus) stimulation of the gastro-intestinal and the genito-urinary tracts, the ovarian and the intestinal mesentery, and the skin causes reflex inhibition of the heart. No reflex cardiac acceleration was obtained in this species.

3. In the turtle reflex cardiac acceleration and rise in blood pressure is produced by the stimulation of the pulmonary and the gastric vagi.

4. In the turtle stimulation of the large and small intestines, the urinary bladder and the cloaca causes, in the non-curarized animal, predominantly cardiac inhibition with fall in blood pressure; in the curarized animals, predominantly cardiac acceleration with rise in blood pressure.

5. In the turtle the spontaneous contractions of the empty stomach do not as a rule stimulate the gastric afferent fibers that act reflexly on the heart or vasomotor centers.

[1] See especially figure 1, *C* and *D*, in that article.

6. In the turtle the stimulation of the central end of the main sympathetic nerves (including the cervical sympathetic) causes predominantly cardiac acceleration with rise in blood pressure.

7. In the turtle visual stimuli may cause reflex cardiac inhibition.

8. In the turtle the discharge of the respiratory center may spread into the cardio-regulative centers, resulting in inhibition or acceleration of the heart. This spread of the respiratory impulse depends on the intensity of the impulse and the excitability of the cardiac centers.

9. In the turtle the cardiac inhibitory mechanism is in tonic activity. This cardio-inhibitory tonus is increased by asphyxia, and may in this condition exhibit a rhythm. The peripheral cardio-inhibitory mechanism is also stimulted by asphyxia.

Note: The tracings reproduced in this article are reduced $\frac{2}{3}$ to $\frac{3}{4}$ of the originals.

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THE RESPIRATORY QUOTIENT AND ITS UNCERTAINTY

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The common definition of the respiratory quotient, i.e., the ratio of the volume of the CO_2 produced to the volume of O_2 consumed $\frac{(CO_2)}{(O_2)}$, is the correct expression when applied to the gas exchange which takes place when a definite substance or mixture is completely oxidized, but appears wholly inadequate when its application is to include the gas exchange of all living organisms or animals. Upon the definition or interpretation of the term will also naturally depend what method may be used in the determination of the respiratory quotient. If we understand the term "respiratory quotient" to apply to the collective exchange of gases due to the metabolic activity of the individual cells forming the body tissues of an animal, then the method for its determination must necessarily be such as to be able to meet the requirements arising because of the nature and complexity of the animal body.

Sources of error. Even among animals of the higher order, like man, having a comparatively simple digestive tract as compared with cattle where the digestive tract is more complex, there is a great difference in the volume, composition and origin of the gases eliminated. Gases from outside and unknown sources may therefore mask the respiratory quotient if the total exchange of gases is the only thing determined. Further, while the total respiratory exchanges for consecutive whole days may show uniformity, through each day there may be periods which differ widely one from another.

Work on human subjects. To illustrate the extent of such variation the author wishes to refer to the elaborate, very complete and valuable metabolism experiments, by means of a closed circuit respiration calorimeter on man with and without food, by F. G. Benedict (1).

From the number of experiments recorded we select and refer to but two of the longer ones—experiments 75 and 76. The first of these

was a fasting experiment, the man remaining in the respiration calorimeter for seven days without food and moving about as little as possible. This was immediately followed by another period of three days in the respiration calorimeter with food, as the second experiment, so that the subject was in the chamber for ten consecutive days. During these experiments the composition of the total ingo and outgo of solids, liquids and gases was determined. The CO_2 production was determined during the entire time for each two hours by absorbing it in soda lime and weighing the amount. Oxygen was supplied from a cylinder which was weighed from time to time, and besides, once each day, the residual air was sampled and the oxygen determined by absorption by potassium pyrogallate. From these weights and determinations together with other data the amount of oxygen consumed during each 2-hour period was computed.

For the seven fasting days the daily 24-hour respiratory quotients agree very well, the average being 0.75, and for the following three days with food the average was 0.817. With the 2-hour periods, however, it is different, for in these we find between the lowest and highest respiratory quotient during the day a difference ranging from 18.84 per cent on one day to as much as 91.83 per cent on another. Stating it differently, the greatest difference between a 2-hour period and the average for the same day was 37.18 per cent. From this we see that if a few small periods should be taken to represent the day, the result may be far from the real average. This is but one of the many lessons we may learn from the valuable experiments referred to.

Again the author wishes to refer to work done at the Carnegie Institution Nutrition Laboratory, Boston, Mass. (2).

To secure the respiratory products in connection with these comparative tests several different methods were used including nose and mouth pieces, masks and bed calorimeter, and the determinations of the gases were made by means of various gas analysis apparatus. The results of a very large number of tests are given. A test period, i.e., the time during which a sample was taken, was usually about 15 minutes except with the bed calorimeter where the periods were 45 minutes. Two to several such short periods taken alternately by the two methods to be compared were grouped together and the averages of such experimental groups are used in comparing the methods.

All these different methods and tests seem to have one thing in common, namely, between successive tests variations in the respiratory quotients obtained were noticed which frequently were quite large.

It is noticed that in about sixty of these groups of tests there was a difference of over 9 per cent between two successive tests or two of the tests within the group, the differences ranging as high as 31.4 per cent between two of the tests in a group. Tests made on the same subject on successive days under similar conditions using the same method of determination, gave respiratory quotients, for the same time of the day, which varied as much as 11.1 to 18.2 per cent for two of three subjects on which such comparisons were obtainable. These tests were made under as ideal conditions as possible for such comparative work and hence we should not expect so much difference.

The frequency of pulse and respiration was given in nearly all cases, but since there does not seem to be any uniform corresponding variation in the pulse and respiration to account for these well distributed differences in the gas exchange, they may, partly at least, be due to causes outside the body tissue. These variations, however, bear out what was said before; that a few short periods can not be relied upon to represent a whole day.

One point to which the author wishes to call attention is that with an animal body, like the human body for instance, we do not deal solely with a complex single unit, but at the same time with the results of the activity of a numberless host of enemies or friends in the form of individuals, mostly microscopic organisms, living within the body yet not part of it.

In man the gases produced by these organisms will vary according to the physical condition of the body as well as the character of the food eaten. Much of the gas produced in the stomach will by means of eructation pass through the mouth and nose whereas that formed in the intestine no doubt will chiefly follow the course of the feces.

In fasting man such gas production would naturally be reduced to a minimum, though not necessarily ceasing altogether, and while some of the variations referred to may be due to the influence of such fermentation gases it is questionable whether, in the case of fasting men, all the variations can be ascribed to that cause. The respiratory quotient obtained on fasting subjects may, because of the variation of such gases, not be wholly applicable to subjects in different states of health and nutrition. Although the total amount of fermentation gases in man is not very large when compared with some other animals, we know that the rectal gases contain considerable CO_2 as well as CH_4. Besides the gases which may pass directly from the stomach to the expired air an unknown quantity of CO_2 undoubtedly is absorbed from the intestinal tract and leaves the body by the way of the lungs.

Work on ruminants. In the case of ruminants the determination of the respiratory quotient is influenced to a very much larger degree by the fermentation gases than with man, for here one-tenth and more of the total CO_2 produced in the body may come from the paunch and intestinal fermentation. Further these 200 to 300 and more liters of CH_4 production per day is a volume large enough to introduce an error in a respiration experiment unless taken account of.

Much work has been done on the respiratory exchange in cattle by the use of respiration chambers but the writer will only refer to the later work of two or three investigators.

W. Klein (3) gives the results of a number of experiments on oxen carried out with the aid of a respiration chamber of the Regnault-Rieset type.

Below is found the result of eight 24-hour experiments with oxen on varying rations using the Regnault-Reiset method.

TABLE 1

Twenty-four hour respiration experiments with oxen

NUMBER	FEED			O_2 USED	CO_2 PRODUCED	CH_4 PRODUCED	RESPIRATORY QUOTIENT	PAGE
	Hay	Chop	Linseed meal					
	kgm.	*kgm.*	*kgm.*	*liters*	*liters*	*liters*		
1	1.5	2.5		2406.0	2371.7	127.0	0.980	203
2	1.5	2.5		2576.4	2407.5	117.0	0.940	204
3	5.0	1.5	1.0	3068.0	2863.0	228.2	0.933	245
4	5.0	1.5	1.0	3021.0	3142.0	244.0	1.040	243
5	5.0	2.0		2606.0	2832.4	158.4	1.087	201
6	5.0	1.0		2610.0	2760.0	146.3	1.057	205
7	8.0	1.5	1.0	3454.0	3548.0	237.0	1.027	241
8	7.0	2.0	25.0*	3727.0	3966.0	316.5	1.064	221
Average of last six				3081.0	3185.2	221.7	1.034	
Average of last five							1.055	

* Beets.

Omitting the first two experiments with the low ration the average respiratory quotient of the other six = 1.034 and the average of the last five = 1.055. These high respiratory quotients obtained during 24-hour experiments by the above method point to something abnormal when we consider the nature and quantity of the food eaten.

Pulmonary respiratory quotient of cattle. To obtain the quotient of the pulmonary respiratory exchange, thirty-two short period experi-

ments of 5 to 23 minutes each were conducted as follows: An ox fed the same daily ration at regular hours was placed in the respiration chamber together with his keeper at varying periods during a number of days. The animal had a cannula fitted into the trachea and by means of a tube was in communication with the outside of the chamber and thus breathed the outside air. Usually several of the respiratory experiments constituting a daily period followed closely upon each other, these daily periods changing so that they would cover and represent the positions and activity of the animal for the whole day.

The tests, made under similar conditions in reference to position and activity of the animal, were grouped together in seven groups, and the average respiratory quotients of these seven groups were 0.816; 0.901; 0.814; 0.849; 0.848; 0.882; 1.003; a difference between the lowest and the highest of 23.2 per cent.

Between the individual experiments making up the groups there was a range of from 0.862 to 0.970 = 12.5 per cent in one, 0.784 to 0.866 = 10.4 per cent in another, and from 0.850 to 0.972 = 14.3 per cent in a third. The greatest difference between any of the tests representing the day was 0.774 to 1.020 = 31.8 per cent. These figures show how great the variation may be in the gas exchange through the lungs with an animal under normal rest conditions and shows again the uncertainty of short period tests. The average of all the tests representing the day was 0.882.

Thus between the average respiratory quotient, i.e., 1.034 obtained with the Regnault-Reiset method and that of the pulmonary breathing, is a difference of 0.152 = 14.7 per cent which is equivalent to 369 liters CO_2. This represents CO_2 which in one way or another escaped from the intestinal tract of the animal. To test this point experimentally Klein made three short experiments of 125, 132 and 119 minutes respectively by placing the animal in the respiration chamber but caused him to breathe outside air by means of a cannula in the trachea, the fermentation products being allowed to accumulate in the chamber. Short respiration tests were made of the pulmonary gas exchange and the residual gases of the chamber were analyzed at the beginning and end of each experiment with the results shown in table 2.

From these short-period experiments the computed CH_4 per day was 150.6 liters. Klein considers this too small a volume and estimates, basing it upon results of other work, that it should have been at least 250 liters. This difference may be due partly to the short experiments,

TABLE 2

Short pulmonary respiration tests

DATE	RESPIRATION PER MINUTE		COMPUTED TO 24 HOURS				
	O_2 used	CO_2 produced	O_2 used	CO_2 produced	Respiratory quotient	CO_2 in chamber	CH_4 in chamber
	cc.	*cc.*	*liters*	*liters*		*liters*	*liters*
6/27/11	2275.5	1972.0	3276.7	2839.7	0.867	393.3	144.0
7/ 4/11	2322.2	1975.0	3344.0	2844.0	0.850	379.0	
7/13/11	2481.7	1908.0	3573.6	2747.5	0.769	411.4	157.3
	2468.2	2140.0	3554.2	3081.6	0.867		
Average.....			3394.9	2866.1	0.845	394.5	150.6

covering perhaps periods of the day of abnormally low fermentation, or else to an excretion of the gases through the lungs. CH_4 having a solubility coefficient equal to 0.035 at 20°C., which is higher than that of O_2, must therefore be absorbed to some extent as well as CO_2 from the paunch where these gases are present in such high percentage. The ratio of CH_4 to CO_2 as found in the above tests was 1:2.62, and therefore the CO_2 corresponding to 250 liters CH_4 would be 655 liters.

On the assumption that the lacking 100 liters CH_4 together with CO_2 at the same ratio as these gases were found in the chamber were excreted through the lungs, then 260.5 liters CO_2 must be subtracted from the pulmonary exchange, and thus the respiratory quotient will be reduced to 0.770. Assuming the other extreme that the lacking 100 liters CH_4 and the corresponding CO_2 were held back or produced after the experiments, then the CO_2 in the chamber should be 654.8 liters for 24 hours. Adding this to the pulmonary CO_2 would raise the respiratory quotient of total gas exchange to 1.037.

That the absorption of CO_2 from the paunch must be much greater than that of CH_4 is self-evident, judging from the very high solubility of CO_2, but the actual amounts of CH_4 and CO_2 which are absorbed from the paunch and the rest of the intestinal tract and pass through the lungs have not been determined.

Ratio of CH_4 to CO_2 in fermentation tests. Markoff (4) has tried to establish experimentally the relation of CH_4 to CO_2 due to fermentation in the paunch and other parts of the intestinal tract of ruminants, by removing some of the content of the various organs and studying the fermentation processes of it while outside the animal body and

gives (4, p. 38) as an average of several very discordant experiments a ratio for CH_4 to CO_2 produced in the paunch of 1:5.19. Gas taken directly from the paunch and analyzed gave (4, p. 2) a ratio of CH_4 to CO_2 of 1:3.68. Thus the difference between 5.19 and 3.68 or 1.51 would represent CO_2 absorbed into the blood. But if CH_4 is also absorbed then the volume of CO_2 passing through the lungs would also be correspondingly larger and the pulmonary respiratory quotient would be lowered accordingly. Applying these ratios to the average respiratory quotient of Klein's six Regnault-Reiset experiments, assuming that no CH_4 was absorbed and lost, the respiratory quotient 1.034 of the total gas exchange will be changed to pulmonary exchange of the following very low values:

With CH_4 to CO_2 = 1:3.68 respiratory quotient 1.034 = 0.768 corrected
With CH_4 to CO_2 = 1:5.19 respiratory quotient 1.034 = 0.660 corrected

Elaborate respiration experiments of 24 to 48 hours duration with a milking cow, breathing through a cannula placed in the trachea, have been made by Holger Mollgaard and A. C. Anderson (5). The incision and insertion of the cannula in the trachea was made in such fashion that in a few weeks the sore was healed and the metal inclosed air tight in the natural tissue, thus permitting the use of the cannula uninterruptedly for weeks at a time without any inconvenience to the animal. These long respiration experiments with cattle are a great improvement over the shorter ones of Klein.

The results are given of one such experiment on a milking cow including two 48-hour respiration tests, during which time the pulmonary gas exchange was determined as well as the CH_4 and CO_2 fermentation products which were accumulating in the respiration chamber.

For the oxygen determinations a new method was used which consists in burning the oxygen to H_2O in a hydrogen flame. While the method is not recommended for short periods the inventors claim that for large samples and long continuous periods it has proved very satisfactory. The air in the chamber was sampled at the beginning and end of a period and the gas analyses made on a Bohr-Tobiesen gas analysis apparatus. The pulmonary respiratory exchange for 24 hours was (5, p. 133) 2762 liters O_2 used and 2670 liters CO_2 produced which equals a respiratory quotient of 0.967. Gases due to fermentation equal 242 liters CH_4 and 681 liters CO_2 in 24 hours, thus the ratio of CH_4 to CO_2 being 1:2.81. In computing the energy production Mollgaard and Anderson make use of the respiratory quotient and claim very satisfactory results.

Applying to the pulmonary gas exchange the 681 liters fermentation CO_2 we get a respiratory quotient for the total gas exchange of 1.213.

In Klein's Regnault-Reiset experiments referred to, one point was not determined which introduced an uncertainty in the respiratory quotient, namely, the amount of fermentation CH_4 and CO_2 which is absorbed in the body and escapes through the lungs. Zuntz and Lehmann have found that there is a small loss of CH_4 through the lungs in the case of the horse where the fermentation mostly takes place in the colon and from Klein's experiments with the ox it seems as though considerable fermentation gases could have left the body through the lungs.

The respiration calorimeter in use at the Institute of Animal Nutrition, State College, Pa., is not a closed circuit apparatus, nor have any tracheal respiration experiments been attempted in connection with it. A few years ago, however, it was thought desirable to try to determine the amount of oxygen consumed besides the total amount of CO_2 and CH_4 produced by the animal while in the respiration calorimeter by the use of the principle of the Jaquet apparatus. By applying a factor for the fermentation CO_2 in relation to the CH_4 it would then be possible to compute the true pulmonary respiratory quotient. The value of such a factor is at present not fully determined.

We realized that in order to obtain reliable results on the oxygen it would be necessary to take and analyze large continuous samples of the ventilating air current. Not knowing of any existing suitable method for the purpose, or that one was at that very time being developed in Denmark, the writer devised and in the course of time succeeded in developing a new method which was tested out in the animal nutrition laboratory. Since no description of this method has been published a brief explanation of it is necessary.

The method consists in removing the oxygen from the outcoming air by means of heated charcoal, and absorbing and weighing the amount of gases formed. A 200-liter aspirator is used for sampling and the collection of the nitrogen of the air sample. The air for analysis is first freed from its H_2O and CO_2 by means of sulfuric acid pumice stone and soda lime, after which it passes through a large silica combustion tube which contains of tested charcoal a quantity large enough to take care of over 300 liters air. This tube which is in a vertical position is heated only over a short section by means of a small electric heating unit. The gases formed by combustion, after the water has been removed, are passed through an absorption set consisting of two KOH

bottles followed by two large soda lime tubes and an acid pumice tube. After being freed from all CO_2 and H_2O the gases pass through a second combustion tube filled with CuO heated to redness to oxidize any CO which is present, and then through another set of soda lime and acid pumice stone tubes in which the CO_2 formed from CO is weighed. The nitrogen passes on to the aspirator in which it is measured. In developing the method many difficulties were encountered because of the physical properties of the charcoal in regard to gases, but all were finally successfully overcome, and the method which although in its present form is rather complicated gave good results. The average of three closely agreeing blank tests of outdoor air was 20.927 per cent oxygen, CO_2 free basis = 20.920 per cent O_2 in air.

Thus, counting non-combustible rarer gases as nitrogen, the composition of the outdoor air will be as follows:

	OUTDOOR AIR	OUTDOOR AIR CH_4 FREE BASIS
	per cent	*per cent*
N_2	79.045	79.0465
O_2	20.920	20.9204
CO_2	0.033	0.0330
CH_4 and H_2	0.002	

Only two of the trial tests with animals were free from accidents and thus available for respiratory quotient determination. Without going into details of the method the main data for computation of the respiratory quotient are given below for these two experiments with milking cows during which the oxygen was determined by the charcoal method. Each set of data covers one subperiod of 12 hours between 6:00 p.m. and 6:00 a.m. The animals were fed twice a day with equal portions of the feed at 6:00 p.m. and 6:00 a.m. (Table 3, see page 62.)

Taking into account only the total oxygen used and the CO_2 produced in the chamber we have the following respiratory quotients:

	COW 885	COW 886
	liters	*liters*
O_2 ingoing	44.1558	45.5362
O_2 outcoming	43.7792	45.2522
O_2 used	0.3766	0.2840
CO_2 produced	0.5847	0.4594
Respiratory quotient	1.552	1.6180

TABLE 3

Data for computing respiratory quotients from two respiration experiments

TESTS AND FACTORS	12-HOUR PERIOD	COW 886 3/5-6/18 DAILY RATION: 3.40 kgm. hay 5.08 kgm. grain	COW 885 5/14-15/18 DAILY RATION: 2.11 kgm. hay 3.15 kgm. grain
		liters	*liters*
Respiration calorimeter experiment	Total (ventilation) outcoming air including N_2, O_2 and CO_2 at 0° and 760 mm	472,712.41	480,273.32
	Total CO_2 produced	1,309.519	1,013.66
	CH_4 produced	119.88	92.01
Air analysis by the charcoal method	Sample of outcoming air for oxygen determination gave		
	Nitrogen	166.8410	172.0560
	Oxygen	43.7792	45.2522
	Carbon dioxide	0.5847	0.4594
		211.2049	217.7676
Using outdoor air composition, CH_4 free basis	The volume of N_2 found in air sample represents ingoing air equal to	211.066	217.664
	Oxygen equivalent in ingoing air	44.1558	45.5362

In this work any error appears magnified in the final results but, aside from analytical error, these results point to some other source for CO_2 than the usual catabolism of feed in the animal tissues.

Applying the facts brought out by Mollgaard and Anderson and by Klein, namely, that for every liter of CH_4 leaving the body directly from the intestinal tract there is also a certain amount of CO_2 given off which is due to an anaerobic fermentation process, the above values of the respiratory quotient will be reduced.

The ratio of CH_4 to CO_2 given by Mollgaard and Anderson was 1:2.81 which for 119.88 and 92.01 liters CH_4 would correspond to 336.863 and 258.548 liters CO_2 respectively. Subtracting these volumes of CO_2 from the total ventilation and from the total CO_2 produced, and recomputing the new CO_2 values back to the volume of the air sample we have the prespiratory quotients shown in table 4.

TABLE 4

Respiratory quotient computed using Mollgaard and Anderson's factor for fermentation gases

	cow 886		cow 885	
	Ventilation	CO_2	Ventilation	CO_2
	liters	*liters*	*liters*	*liters*
Total ventilation and CO_2...	472,712.41	1,309.519	480,273.32	1,013.66
Total fermentation CO_2.....	336.863	336.863	258.548	258.548
Corrected total ventilation and CO_2.....	472,375.547	972.656	480,014.772	755.114
Air sample for O_2 det.....	211.066	0.4346	217.664	0.3424
O_2 used.....		0.3766		0.2840
Respiratory quotient.....		1.154		1.210

If instead of the ratio 1:2.81 for CH_4 to CO_2 the value found by Markoff for the gas taken directly from the paunch of an animal, which was 1:3.68, is used, or if we take it for granted that there is an absorption of CO_2 from the alimentary tract and loss of the same through the lungs, and use Markoff's ratio of 1:5.19 for CH_4 and CO_2 due to fermentation and repeat the computation we would have the following:

	RESPIRATORY QUOTIENT	
	Cow 886	Cow 885
CH_4 to CO_2 ratio = 1:3.68.....	1.031	1.078
CH_4 to CO_2 ratio = 1:5.19.....	0.816	0.856

From this present uncertainty about the respiratory quotient we may well conclude that it does not furnish a short cut to the accurate computation of the energy metabolism of the animal. On the contrary we must expect that as the nutrition problems become more and more complex, more accurate, more refined, more comprehensive methods will also be needed and that the research worker must call to his aid not only general chemistry and physics but bacteriology, biological chemistry, etc., to help solve his problems.

Thus one of the fundamental questions in reference to the respiratory quotient determination by the open circuit Pettenkofer or Jaquet and other methods as yet undetermined is that of the ratio of the CH_4 to CO_2 as produced throughout the whole intestinal tract of the animal.

In reference to the pulmonary respiration experiments the important point is to know how much of the fermentation CH_4 and CO_2 is absorbed into the blood and is escaping through the lungs so that proper corrections can be applied for them. Based upon work already done but which at present is by no means perfect some factors could perhaps be agreed upon tentatively and used until better ones are established.

Another question, perhaps not of so much importance yet demanding an answer, is whether the oxygen of the air taken into the digestive tract with the food is absorbed into the blood, or whether some aerobic process is going on simultaneously with the anaerobic, in the digestive tract, thus, in turn, to some small degree at least masking the fermentation process.

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THE RELATIVE HEIGHTS OF THE EATING AND DRINKING ARCS IN THE PIGEON'S BRAIN, AND BRAIN EVOLUTION

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The phenomena produced by cerebral ablation in birds are especially interesting because of the light they seem to throw on the evolution of the brain. In a recent article Martin (1), from his observations on decerebrate chicks, offered the interesting suggestion "that the establishment of function within the sub-cerebral nervous structures took place when life was predominantly marine and when, therefore, deliberate drinking was unnecessary," hence it might be inferred that there is no sub-cerebral mechanism for deliberate drinking. On the same grounds we might infer that, since deliberate feeding must have been already established, there is a sub-cerebral mechanism for deliberate feeding. Then when some of the descendants of these animals began to spend much or all of their time on land, and deliberate drinking became necessary, the mechanism for deliberate drinking had to develop in that portion of the brain which was still developing, hence in the cerebrum.

But some *recent observations of mine on decerebrate pigeons* appear to lead to different conclusions, as the following account will show. On June 10, 1920, an attempt was made to remove all of the cerebral hemispheres from pigeon 3, without injury to the other parts of the brain. At the end of the experiment, the necropsy showed that this had been practically accomplished. Most of the cerebral tissue that remained was yellow and hard. There remained only a few diminutive, convolution-shaped structures of the base of the cerebrum that could possibly have remained capable of functioning. (Whether or not they were actually functional, we hope to determine by microscopic examination.) The parts of the brain below the cerebrum had not been touched. The pigeon appeared to recover quickly. No

outward evidence of infection was seen, unless an increase in the respiratory rate that lasted only a part of the second day could be evidence of infection. There had been no more than a transcient trace of discoloration or swelling about the eyes. The only thing noticed at any time that might have had an influence on the health of the bird, was the fact that the intestine contained some worms. This was discovered on the 14th day after the operation, when a few worms were passed, and the necropsy showed worms in the intestine. It is not thought that this had any material influence upon the return of reactions.

In this pigeon *spontaneous drinking activities* returned. They returned earlier than spontaneous eating activities. In fact, it was only two and a half days after the operation that it showed effective drinking, but it never showed effective eating. At this time it showed such spontaneous drinking as others have noted in decerebrate birds; that is, when its bill was brought into water, it drank. This was brought about in this case by tilting the bird toward some water in a pan resting on the table. The bird had been weighed before being tilted. After two or three tiltings and drinkings of this sort, it was dried and weighed again, and was found to have drunk 5 grams of water. This excess of drinking over eating could hardly have been due to an excess of thirst over hunger for, on the day before, it had been given 40 grams of water by pipette, while it had been given only 10 grams of grain and had vomited all that. (This drinking reaction might possibly have been obtained earlier, had it been tried earlier.)

Although others had observed the kind of spontaneous drinking just described, I am not aware that others had noted "any suggestion of a *deliberate attempt to obtain water.*" But this bird, on the 6th day after the operation, gave the appearance of a deliberate attempt to obtain water in that, when he was tilted only enough to bring his beak near the water, he "deliberately" or spontaneously thrust his beak in and drank. Just how much deliberation there was in this attempt it is, of course, impossible to say. It may be that under such circumstances there would not be much deliberation in the attempt of a normal pigeon to obtain water. Be that as it may, this decerebrate pigeon appeared to act exactly as a normal pigeon sufficiently tamed might have acted under the circumstances. Again on the 18th day there was the same sort of "deliberate" drinking. It was also noted at this time that the bird appeared interested in the water. The sight-touch-drinking reaction was sufficiently efficient to furnish the bird all the water it needed for maintenance; for, from the 15th to the 35th day inclusive

the bird received no water other than that taken spontaneously. And during these twenty-one days the pigeon remained strong and healthy and its weight was maintained; and at times when the grain was sufficient the weight even increased.

The *"feeding activities" during the first twenty-six days* are in marked contrast with the drinking activities, which have just been described. Never during this whole period did I succeed in getting the pigeon to peck at a grain except once, feebly and wholly ineffectually. This occurred on the third day when a grain was rubbed lightly against the beak. Nor was it ever seen to peck at any other objects in this period, or even to show an inclination to peck. Daily attempts were made to bring about this reaction and several different methods were employed, but absolute failure was the uniform result. Day after day grain was sprinkled on the table about it. Grain was held up to it in a small pan and even rubbed gently against its beak. The pan was shaken so that the sound might aid sight in bringing about a reaction. (Contrary to the impression conveyed by the literature, these decerebrate pigeons hear well and react differently to different sounds.) The pigeon was put into its old cage with its former companions, which it had uniformly dominated for the four months preceding the operation, and with which it had eaten during more than those four months. Here grain was scattered about it with the hope that the example and sound of the pecking of its old companions would bring about the reaction. This was repeated day after day, but in vain. At times grain was held up to it in a pan or in the hand while a companion ate out of it. This produced no feeding reaction. Starvation for forty-four hours brought none. A diet low enough to reduce its weight to 211 grams (its normal weight was about 300 grams) did not bring it into a condition that would cause a feeding response. Nothing that I could think of succeeded in bringing about in this period the simplest pecking reaction, with the single exception of the slight movement mentioned above. Hence there was almost an absolute absence of eating activities in this period of twenty-six days, in which there were spontaneous drinking activities sufficient for maintenance.

Of the other various activities that have heretofore been regarded as feeding activities it showed only one, viz., the swallowing reflex, elicited by dropping grain well back into the mouth. (Grains placed in the front part of the beak were thrown out.) It did, however, occasionally show another activity that might possibly be an infantile feeding reaction. This activity was a kind of "gaping" that has been seen in

decerebrate pigeons by others. It was so related to hunger and to external food stimuli as to suggest that it might be related to the infantile reaction for the reception of food.

We see then that during the first twenty-six days not only was deliberate eating absolutely absent, but there was manifested only a trace of any feeding activity other than the swallowing reflex. Thus the observations of the first twenty-six days appear to indicate that the removal of the cerebrum really injures the feeding mechanism much more than the drinking mechanism, and it will be seen from what follows that the observations in the succeeding twenty-three days, i.e., to the end of the experiment, appear to confirm this conclusion.

During this *succeeding period*, the efforts to increase both the eating and the drinking reactions were continued. As was stated above, in this period up to the 36th day, no water was poured into the beak. During this portion of the period, as in the period from the 15th to the 26th day, the bird was induced to take enough water spontaneously to maintain its weight and strength. This was always brought about by tilting the bird, or by raising the water to or near to its beak, with two notable exceptions. On these two occasions the bird showed perfect independence in drinking. These two responses I will describe in some detail, not only because they show an efficient sub-cerebral drinking arc, but also because, so far as I am aware, they have not before been noted in decerebrate animals.

The first absolutely independent drinking occurred on the 32nd day and was brought about as follows: The pigeon had begun showing the pecking reaction on the 27th day and on the morning of the 32nd day it was placed near the pan of water as usual, and the pecking reaction was induced by holding grain up to it in a small scale pan. The pan was gradually lowered and the pigeon was soon induced to peck at grains scattered on the table. When the pecking showed signs of ceasing, it was reinduced by stirring the grains near by with the finger. In this way the pigeon was kept pecking at grains on the table for some fifteen minutes with brief intermissions. In this time there was some walking about in the neighborhood of the pan of water. (In the fifteen minutes of pecking the pigeon was not seen to get a single grain into the beak.) Suddenly it was seen to start directly toward the pan of water. (The pan containing the water was the same one that it had drunk from for nearly five months before the operation. This might possibly have had some influence on the drinking reaction, but probably had none.) When he reached the pan he stepped up onto its

edge, balanced a little, and made some motions with the beak toward the water. He then stepped down, pecked once or twice at the water, then "deliberately" put the beak in deeply and appeared to drink well. He then withdrew the beak normally, then, without moving from his place, he repeated the drinking once or twice. The bird was dried and weighed and found to have drunk one and a half grams of water. I was surprised for, in my experience, this amount of water is a small drink for a pigeon, and the drinking had occupied much more time than a normal pigeon would have been likely to use under the circumstances. But it showed definitely a wholly efficient and independent drinking reaction in a decerebrate animal. This reaction also comes pretty close, in outward appearance, to an act of "associative memory." It simulated also deliberation, choice, an "act of will." It showed that there was, intact within the part of the brain below the cerebral hemispheres a moderately efficient sight-drinking arc. But the fact that the bird even when very thirsty (deprived of water for twenty-four hours or more) did not often show this independent sight-drinking, suggests that the sight-drinking mechanism had been partly taken away when the cerebral hemispheres were removed. We may infer that, although the decerebrate pigeon has a pretty good sight-drinking arc left, the sight-drinking mechanism of the normal pigeon is spread partly in the cerebral hemispheres and partly in the subcerebral brain.

The second reaction of independent drinking occurred on the 35th day, as follows: On this morning in his walking about, pecking at grains and occasionally at other things, from time to time he wandered near the pan of water, and sometimes appeared to look at the water, but not attentively. I put my finger into the water near the edge of the pan opposite to him and agitated the water a little. This appeared to attract him almost immediately. He walked up to the pan and after a little hesitation pecked once in the water, then almost at once put his beak into the water normally, drank a little, took his beak out and raised his head, then immediately put his beak back into the water and drank again. Unfortunately he had not been weighed and therefore I was unable to determine how much he had drunk.

There were other occasions when the pigeon was seen to be independently attracted by water and even to deliberately peck in it but without drinking. On another occasion, the 44th day, he was left to himself for about two hours. He had been placed on the table about 45 cm. from a small beaker of water standing not far from the pan of

water. When I returned I found the beaker upset, the water spilled and the bird resting in the middle of the water pan, belly-deep in the water, evidently contented.

All this suggests that if sufficient care were taken there is a possibility of obtaining a decerebrate pigeon that will drink, absolutely independently, sufficient water for maintenance. We may infer, in any case, that there is a fairly efficient drinking arc intact in the subcerebral brain of the pigeon.

It was different with the feeding activities in the last half of the experiment. No independent feeding could be induced, but the feeding activities increased markedly over those of the preceding period. Beginning with the 27th day there was a rapid increase in the amount and vigor of pecking, and in the walking about over the food with the air of seeking something to eat. In the later days there was at times considerable cooing over the grain. On the 27th day when grain was held up to him in the scale pan, he looked down at the grain and made two or three slight movements with his beak toward the grain. About half an hour later when the test was repeated, he made three short pecking movements toward the grain. The movements appeared to involve considerable effort. They were tremulous, and suggested vomiting movements almost as much as pecking movements. Moreover, they reached only half way to the grain which was about two inches away. At the next trial he made five pecking movements. Finally he succeeded in striking the grain. This was twenty-one days after the first deliberate drinking. On the next day, at the first trial, the pecking movements were of much the same character, but later in the same day he pecked the grain in the pan repeatedly and vigorously, both alone and later with a companion. The day following, the 29th, he continued thrusting his beak vigorously into a beaker of grain for some time, flirting out much grain. A little later he pecked three times at my finger, this being the first time he pecked at anything other than grain. On the 32nd day, as stated above, he walked about for more than fifteen minutes, pecking at grain scattered on the table, but apparently he got no grain into his beak. He was encouraged in this by my occasional stirring of the grains near by with the finger. In the succeeding days the feeding reactions were of much the same character. Some days he was more reactive, some days less. On the 35th day he was easily kept pecking for more than forty minutes. This was the maximum feeding activity in the forty-nine days of the experiment. Sometimes, even though probably hungry, he would not peck at all.

In spite of all this pecking in the last twenty-three days, so far as I could ascertain, he never got but one grain into his beak, and he dropped that. Although at times it appeared that he might possibly have eaten some grains, I could not convince myself that he had. For example, on the 37th day (the day when he was seen to get a grain into his beak and to let it drop), it was thought that he might possibly have eaten some grains. So he was weighed after the active pecking ceased, and was found to weight one-tenth of a gram less than he had weighed about a half-hour earlier (allowance being made for feces passed in the meantime). In order to be sure whether this indicated eating or not, it would be necessary to know how much weight he lost in the time through the exhalation of carbon dioxide and the evaporation of water. I therefore attempted on the 40th day to determine the normal loss of weight exclusive of feces. He was found to lose in this way, four-tenths of a gram in thirty-four minutes. This looked as though he might have eaten some on the 37th day, but it was not absolutely convincing, and observations on other pigeons make it look doubtful. It appears therefore that the decerebrate pigeon is practically incapable of seizing a grain with its beak. The chain of reactions which together constitute eating, appears to be broken at this point; the reaction for seizing grain appears to be missing.

Moreover when grains were placed in the anterior portion of the beak, this bird uniformly threw them out, although grains dropped well back into the mouth were as uniformly swallowed. Here then there appeared to be another link missing, viz., the reaction for moving objects from the anterior portion of the beak to the posterior portion.

Since, as the experiment progressed, there appeared no convincing evidence of the reappearance of these two missing reactions, one is tempted to conclude that the corresponding centers had been removed when the cerebrum was removed.

This experiment suggests, then, that one or two correlation centers absolutely necessary for successful feeding in the pigeon lie in the cerebrum. And when the cerebrum is removed, spontaneous feeding becomes impossible because these indispensable centers are taken away. In other words, the removal of the cerebrum leaves the sight-eating mechanism irreparably broken. It suggests also that although a portion of the sight-drinking mechanism lies in the cerebrum, there still remains, after the cerebrum has been removed, an intact sight-drinking arc in the sub-cerebral brain; and that this sub-cerebral drinking arc is capable of bringing about wholly independent and efficient drinking

The question arises, will decerebrate pigeons in general, under favorable circumstances, react as this pigeon reacted, i.e., in such a way as to indicate that pigeons have a fairly efficient nervous mechanism for drinking below the cerebrum, while their nervous mechanism for eating is connected up only in the cerebrum?

A study of two other pigeons appears to confirm these conclusions. One of these pigeons, no. 2, a hen pigeon, was kept healthy and under observation for nearly five months after the operation, which was performed on January 21, 1920. (It had had semicircular canals operated upon the day before for class demonstration.) Necropsy showed that practically all but not quite all of the cerebrum had been removed and, in addition, a thin slice off the top of the brain stem (which includes the thalamus in the pigeon), leaving the third ventricle opened up from end to end, and laying bare the posterior commissure. The surface of the portion of the brain remaining at the end of the experiment was smooth, clean, fairly symmetrical and of normal color, hence it was inferred that there had been no inflammation after the operation. Owing to press of other work, the study of this pigeon was not as thorough as that of no. 3, but the observations made appear to confirm those made on that pigeon. In this pigeon there was less of both eating and drinking activity than in no. 3, although the walking and the cawing activity in the last months of this experiment were much greater. While both the eating and the drinking mechanisms appeared to be more injured than in no. 3, in this pigeon also, the eating mechanism appeared to be much more seriously injured than the drinking mechanism.

Drinking activities were not seen in this pigeon until comparatively late. Attempts were often made to obtain eating and drinking reactions. I did not see any semblance of spontaneous drinking until the 51st day, when it appeared to drink spontaneously from a beaker resting on the floor. The beaker of water had been placed on the floor of its cage after ineffectual attempts to get it to drink by raising the water up to its beak, and I had withdrawn to a distance of about 5 feet. It took a step or so toward the water, put the beak down directly into the water and appeared to drink. Evidently the failure to drink when the water was raised to its beak was not due to absence of the necessary mechanism, nor to absence of thirst (i.e., the need of water), but to some *inhibition*, due perhaps to my proximity. It usually turned away when water was raised up to it, and sometimes took a few steps away from the water, not, however, showing any marked

signs of alarm. Sometimes after the beak had been wetted by the raised water, this, instead of bringing about the drinking reaction, caused a shaking of the head as if a disagreeable sensation had been felt. On the 145th day it spontaneously thrust its beak deep into the water, but appeared to withdraw it without drinking, although it made slight movements of the beak afterwards that might have been drinking movements. The many other attempts both by myself and by another capable observer, during the almost five months, failed to bring about in this pigeon any drinking activity other than occasional slight movements of the beak after it had been wetted.

The observations in this case, then, *indicated* serious injury to the drinking mechanism but not sufficient to break it entirely. The one instance of spontaneous drinking appears to show that the sight-drinking mechanism was still sufficiently intact to bring about an efficient reaction when the conditions were sufficiently favorable. In other words, in pigeon 2 as well as in pigeon 3 there was an intact drinking mechanism in the subcerebral brain.

Of *feeding reactions* there was still less evidence. In spite of the fact that in the later months when hungry it walked about constantly, cawing much of the time, I did not in the whole five months see it peck, except on one occasion, and then not at the grain held up to it. It was on the 139th day. Grain was held up to it, and two different times it turned away from the grain and pecked at the white oilcloth upon which it was standing, each time in about the same spot, a little to the right and in front of its feet. There was no speck on the oilcloth near the place of the pecking. (This reaction might have been modified by injury to the semicircular canals by the operation on January 20, 1920.) Gaping was seen as early as the 3d day, and a reaction suggesting awkward ground-scratching on the 5th day. It first got out of its box and walked about on the floor on the 11th day, apparently driven by hunger. After a few weeks the walking and cawing appeared to occur regularly every time the bird got hungry. Absence of food and water for several days at a time failed to bring about any other feeding or drinking activities. Grains placed in the anterior portion of the beak were thrown out, as in the case of no. 3.

The observations, therefore, over nearly five months show no evidence of an intact sight-eating mechanism in no. 2. The eating mechanism appeared to be even more seriously injured than in no. 3. The little eating and drinking activity of this bird compared with nos. 3 and 1, suggests that the removal of a thin slice from the top of

the brain stem of a decerebrate pigeon produces further injury to both eating and drinking mechanisms.

In the case of pigeon 1, the conditions were different but the indications in regard to eating and drinking practically the same. The pigeon was operated on January 22, 1919. There appeared to be enough inflammation from the operation to cause some discoloration and swelling about the eyes and to disturb the equilibrium a little, but on the 6th day the pigeon appeared well. Necropsy showed more cerebral tissue remaining than in the other two cases, especially in the frontal region. The sub-cerebral portion of the brain had not been touched. The pigeon remained healthy and was under observation for almost twelve months after the operation and then met accidental death.

The eating and drinking activities in this pigeon were scarcely greater than those of no. 3, and much slower in making their appearance, in spite of the fact that he had more brain remaining. This slowness might possibly have been due in part to the mode of procedure in trying to induce the reactions. As with nos. 2 and 3, in this case also the drinking activities were markedly more efficient than the eating activities, indicating, here also, more injury to the eating mechanism than to the drinking mechanism.

Spontaneous drinking reactions returned gradually. They first made their appearance on the 36th day after the operation when, after the water was raised to wet his bill, he thrust his beak in and drank. On the 38th day he thrust the beak in when the water was only brought near. All the water he got in the ten months from the 57th day until his death, he took spontaneously. On the 57th day he moved his beak down about four inches to get it into the water. On the 58th day, when the beaker of water was held beside his tail he turned and put his beak down into the water and drank. On the 69th day and thereafter he drank from the beaker resting on the table, but usually held by the hand. To initiate the reaction, it was usually necessary to slide the beaker on the table. On the 135th day he took two or three steps toward the beaker, put his beak down into the water and drank, but it was not until the 153d day that he again walked spontaneously toward the beaker. On the 160th day he returned his beak to the water twice without urging (i.e., without having the beaker moved in the meantime). On the 173d day he turned a little, walked five steps and put his beak directly into the water and drank 1.8 grams without a false move. On the 177th day he followed the beaker a few

steps when it was slid away from him. On this occasion he repeated this act several times in succession. His drinking reactions continued about like this until the 356th day, the day of his death. On some days he was more responsive and on others less.

The *quantity of water taken* spontaneously appeared to be related to the quantity of grain given. When more grain was given more water was taken spontaneously. It also appeared to be related to the surrounding temperature and humidity conditions. In mild weather, with an ample ration of grain the amount of water taken was about 75 per cent of the weight of the grain. But in hot weather, when the grain ration was small it rose to 167 per cent of the weight of the grain. In one week in March the average daily quantity of water taken was 8.3 grams; in a week in April it was 15.9 grams.

We have here, then, evidence of an intact and fairly efficient sight-drinking mechanism.

The return of feeding reactions other than swallowing and "gaping" was also gradual. Pecking was first seen on the 9th day when there was feeble and uncertain pecking at a grain of corn rubbed against the beak. On the 40th day he pecked at the table top. On the 61st day he pecked at grain on the table, but got none into his beak. On the 85th day he cooed on seeing and hearing grain sprinkled on the table before him. On the 97th day, awkward ground-scratching was first seen. Later it became more pronounced and more frequent. Still on the 153rd day he pecked at grain only rarely; 161st day pecked at grain in the beaker; 163rd day, pecked at grain more than usual, got a grain of rice into his beak but dropped it; 174th day, got two different grains into the beak but dropped them both; 181st day, got a grain of wheat into his beak and held it a moment, making what appeared to be testing movements, but soon he dropped it. From this on to the day of his death, there was no further increase in his eating activities, in spite of the fact that some other responses, not bearing directly on the question under discussion, showed a continued increase in sensitiveness and in reactivity. No actual eating took place, although the pigeon remained alive and healthy for nearly twelve months after the operation. Grains put into the anterior portion of the beak were always promptly thrown out. Hence it appears that the nervous mechanism for feeding in this pigeon was in about the same condition as that in pigeon 3. The same two links were missing, and failed to reappear in twelve months' time, under what might be regarded as fairly favorable circumstances.

Hunger was never seen to cause any of these birds to fly.

DISCUSSION

The fact that in all these birds, i.e., in all the birds I have studied thus far for these reactions, the chief phenomena bearing on the questions under consideration recur with such uniformity in the different birds, suggests strongly that further study would only confirm these findings, and that they are characteristic of the decerebrate condition in the pigeon. The fact that all three of these birds showed independent and spontaneous drinking reactions, indicates pretty clearly, that there is a fairly efficient sight-drinking arc in the sub-cerebral brain of the pigeon. (This part of the pigeon brain includes the thalamus but not the corpus striatum.) This fact that the fundamental drinking mechanism is practically complete below the cerebrum implies that in the history of the race, the formation of an efficient drinking mechanism, and the generation of the reaction of *deliberate drinking*, antedated the development of the cerebrum. The fact that the removal of the cerebrum produces a rather marked change in the reactions to thirst and to water indicates that when the cerebrum is taken away, some of the drinking mechanism as it exists today goes with it. That is, these are indications that, while the ancient drinking arc lies below the cerebrum, it possesses accessory arcs or shunts that are spread up into the cerebrum; that, therefore, there is an ancient drinking center in the sub-cerebral brain, and one or more modern drinking centers in the cerebrum.

The fact that it appears impossible to get any of these decerebrate pigeons to actually eat a single grain, indicates forcibly that the removal of the cerebrum disrupts the nervous mechanism for eating irreparably. The fact that these birds appeared never to get a grain into the beak except by accident suggests that the center for seizing with the beak had been removed; that is, that this particular eating center lies wholly in the cerebrum. The fact that none of the grains placed in the anterior portion of the beak, nor any of those seen to be gotten into the beak by accident when pecking, were ever moved to the back portion of the beak, appears to justify the conclusion that the correlation center for moving objects from the anterior portion of the beak to the posterior portion had been removed, i.e., it too lies in the cerebrum. The fact that grains placed in the anterior portion of the beak were uniformly and promptly thrown out of the beak shows that a good-working correlation center for this act lies in the sub-cerebral portion of the brain. It also suggests that the correlation

of taste with swallowing was brought about in the cerebrum, and that the taste center lies in the cerebrum only. The fact that no matter how long these birds were starved, even to the extent of several days, none of them was ever seen to fly to seek food, indicates that the center which correlates flying with hunger lies in the cerebrum. Under suitable conditions hunger in these decerebrate birds was followed by walking, cawing, cooing, pecking, flirting of grains with the beak, and ground-scratching, also by looking about. This proves the presence of the corresponding correlation centers, i.e., that they lie in the sub-cerebral brain.

These birds, at many different times, walked toward water or grain. Therefore, there are mechanisms below the cerebrum that mediate attraction. Often these birds would turn their heads away from offered grain or water, hence there are also centers below the cerebrum that mediate repulsion. These two classes of phenomena indicate that the decerebrate pigeon is not indifferent to its environment, or that its environment is not indifferent to it. They also show that decerebrate pigeons do not react fatally, i.e., always in the same way to the same stimuli; that is to say, they are not like automata, but show a certain range of "choice." The fact that no. 2 on one occasion, though thirsty, did not attempt to drink when water was held up to it, but did drink when the water was set down and the attendant retired somewhat, showed that there is at least one mechanism for inhibition intact in the brain below the cerebrum. (The presence of other inhibitory mechanisms was shown by other phenomena, observed but not bearing directly on the subject of this paper).

The fact that the feeding and the drinking reactions often reappear late and continue to improve for some time proves that the return of certain functions after cerebral ablation does not occur suddenly, but is a gradual process. In spite of the slowness of this recovery, on account of what is known of regeneration and growth in the central nervous system, it can hardly be inferred that new structures were produced in the sub-cerebral brain for these reactions. Nor does it imply that "structures in the lower part of the brain take over the functions of structures in the cerebrum." The legitimate conclusion seems to be that the connections in these particular old arcs are not thoroughly patent in the normal pigeon, but under favorable circumstances they become more and more patent for a considerable time after the cerebrum is removed, thus finally permitting the impulses to pass more freely and the reactions to take place. It also implies that

in the normal bird these particular reactions are almost wholly guided by arcs extending into the cerebrum; that the accessory arcs to these ancient mechanisms have now become the chief arcs and that the primitive chief arcs in the sub-cerebral brain are now in the process of breaking down or becoming blocked.

These results in the light of our present knowledge therefore suggest *an explanation for the fact that the nervous mechanism for eating is connected up only in the cerebrum* in the pigeon, while there is a drinking mechanism fairly complete in the sub-cerebral brain. It appears to be because the stimuli of two essential eating reactions have constantly changed from the earliest times to the present day, while the stimulus to the drinking reaction has always remained practically the same.

The *two eating reactions whose stimuli have changed* are: first, the seizing of food with the beak; second, the propulsion of the seized food to the pharynx. The food of the earliest ancestors of the pigeons must have been very different from that of their descendants, the pigeons of today. For example, when the ancestors of the pigeons lived wholly in the water, it is inconceivable that their food could have been grain. It seems perfectly safe to assume that at that date the seizing of food was incited by the sight of those objects which they had become accustomed to eat. When one of those prehistoric food objects came into the range of vision of a hungry ancestor, it acted as a stimulus that set up the reaction that resulted in the seizing of it. As soon as the seizing had taken place, another reaction was set up through a taste arc. If the taste was good the resulting reaction was a propulsion of the food toward the pharynx; if the taste was very bad or absent the reaction might well have been a throwing of the object out of the mouth. With changes in environment some of the old food objects ceased to come into view, therefore the particular arcs which had served to bring about their seizing fell into disuse. This may have caused atrophy of the arcs and finally dissolution or at any rate a disappearance or a blocking of the synapses. But the change of environment brought into view new food objects and sight of these produced new seizing arcs in that portion of the brain which was still plastic. New changes of environment, in a similar way, brought about the disappearance of other old food-seizing arcs, and the formation of other new ones in the plastic region of the brain. The taste arcs that initiated the beak-to-pharynx-propulsion reflex underwent similar changes. The disappearance of old tastes caused the disappearance of the corresponding taste arcs. The stimulus of new food tastes caused the formation of corresponding

new arcs, that initiated the propulsion toward the pharynx. Thus, as time went on, the connections of the old food-seizing and the old food-testing arcs were constantly dropping out of the old brain, and new corresponding connections kept forming in the new brain until, finally, none at all were left in the old brain. And so, today, food-seizing and food-testing correlation centers are found only in the cerebrum.

But with *the drinking reactions and arcs* it was different. Water is today practically what it was in the beginning, in so far as the pigeon is concerned. Light from water has ever struck the retina in about the same way, and the impulses so produced have passed over the same arcs, to arouse practically the same drinking reaction. On this account the essential sight-drinking arc has remained much as it was in the beginning. It therefore, became fixed relatively early and remained a practically unchanging part of the ancient brain. In the later development, while the cerebrum was being formed, there was no force acting to change this primeval structure except to produce side arcs that rendered the reaction more prompt to the needs, and better adjusted to the comparatively slight changes that had taken place in its conditions. Thus we have a plausible explanation of the presence of a pretty efficient drinking arc in the sub-cerebral brain.

These results then, in the light of our present knowledge, suggest that the *chief causative factors in brain evolution* are use and disuse, and reactions upon the arcs themselves through arcs that return to them.

The *laws of cerebral evolution* may be tentatively stated as follows:

1. Moderate *stimulation* improves the nutritive condition of a nerve cell and while the cell is still plastic thus tends to produce arcs and reactions.

2. *Use* tends to keep an arc efficient.

3. *Disuse* tends to block an arc or to break its connections.

4. *Return-arcs* bring about return-stimulations or return-inhibitions of an arc, depending upon the effect of the reaction, whether favorable or unfavorable. For example, seizing a good grain would be a favorable reaction, a taste arc that would bring about the seizing of another would be a return-arc, bringing a return-stimulation. In a similar way seizing a very bad grain, an unfavorable reaction, would bring about a return-inhibition.

5. The effects of *return-stimulations* on an arc are improvement of its nutritive condition and wider opening of the involved connections. Therefore uniformly favorable reactions tend to become quick and

strong, and they tend to make their arcs stable and thus to establish more permanent portions of the brain.

6. The effects of *return-inhibitions* on an arc are a depression of its nutritive condition, a diminution of the patency of the involved connections and an opening up of new connections to produce new substitute arcs. Therefore, uniformly unfavorable reactions tend to destroy their arcs and to set up substitute arcs in their stead.

7. The effects of *alternations of return-stimulations and return-inhibitions* on an arc, are the preservation of a moderate nutritive condition in it, the holding of the involved connections moderately open, and the favoring of the formation of new connections to produce alternative arcs or shunts. Therefore, arcs yielding both favorable and unfavorable reactions tend to spread by shunts into the newer, more plastic portions of the brain, and thus to form broad or deliberative arcs.

These laws are not only laws of brain evolution; they are also *laws of education*, because education consists in the formation of new connections for the production of new ideas, and the inhibition of old connections that produce ideas not in accord with the facts.

The findings suggest also a re-statement of the *theory of the functions of the cerebrum, in contradistinction to the sub-cerebral brain*, or rather, of the new brain in contradistinction to the old.

They appear to suggest that in the pigeon the *special function of the new or plastic brain* is the formation of new connections, and the mediation of those reactions that are relatively new and of those that are prone to result unfavorably or that are fraught with inhibitions. The distinguishing characteristics of cerebral reactions are inhibition, deliberation, judgment, choice, will, new adjustment and new initiative. Inhibition involves delay of reaction and results in deliberation, i.e., the passing of the impulses into alternative arcs and lightly to the corresponding effectors, so producing the anticipatory feeling of the corresponding reactions and the imaging of their probable results. Thus deliberation, at least in its higher forms, yields judgment, i.e., the feeling that the reaction of *one* of these alternative arcs would be the most favorable and so judgment yields choice and will, i.e., the throwing of the impulses through the chosen arc. In a similar manner, unfavorable judgment of all the alternative arcs, i.e., the feeling that the reaction of neither would produce a favorable reaction may, when the cells are still plastic, yield new connections and so new arcs and new reactions, or new initiative. Therefore, the cerebral reactions bear the especial stamp of intelligence, and the cerebrum is the chief seat of learning.

On the other hand, the *special function of the old or fixed brain and cord* is the mediation of primitive reactions that have been long wont to result favorably. In the pigeon they are especially the visceral reactions, reactions of equilibrium and of locomotion, and certain preservative reactions such as retraction from noxious stimuli and self-cleaning reactions. But, under favorable circumstances, it is also capable of mediating certain hunger, thirst and sex reactions, and other preservative reactions, since certain of its primitive functions, which in the course of time have partly or completely passed over into the cerebrum, may be regained after cerebral ablation, if the circumstances are sufficiently favorable. Thus it may mediate certain initiatives and inhibitions, and it gives at least semblances of mediating forms of deliberation, choice and will. It mediates forms of memory in so far as they are necessarily involved in the mediation of the above reactions. The extent to which sensation is mediated by the sub-cerebral brain can be only vaguely inferred as yet, but it seems that the semblances of deliberation, judgment, choice and will that are manifested imply that there may be present with these phases of the reactions at least *some* vague, corresponding sensations. Hence the sub-cerebral reactions, actual and possible, have the character; some, of the reflexes; some, of the simpler instincts, and in this old portion of the brain learning is at a minimum.

CONCLUSIONS

A study of the eating and the drinking reactions of three pigeons after cerebral ablation, one for seven weeks, one for nearly five months, and the other for almost twelve months, appears to lead to the following conclusions in the case of the pigeon:

1. There is a fairly efficient sight-drinking arc in the sub-cerebral brain (which in the pigeon includes the thalamus but not the corpus striatum).
2. The quantity of water taken spontaneously by a decerebrate pigeon under favorable circumstances, depends upon the quantity of grain given, and is well adapted to the needs of the organism.
3. In the history of the race, the sight-drinking reaction antedated cerebral development.
4. The normal nervous mechanism for drinking is broad and has spread into the cerebrum.
5. The removal of the cerebrum disrupts the nervous mechanism for eating irreparably. In other words, it breaks the chain of reactions

essential to actual eating by removal of at least two essential correlation centers.

6. The correlation center for seizing with the beak lies only in the cerebrum.

7. The correlation center for the propulsion of objects from the anterior to the posterior portion of the beak lies only in the cerebrum.

8. In the absence of taste or other sense to initiate the beak propulsion reaction, a beak-ejecting reflex through the subcerebral brain, throws all objects out of the anterior portion of the beak.

9. The taste center probably lies in the cerebrum only.

10. The correlation center for hunger-flying probably lies in the cerebrum only.

11. There are correlation centers for hunger-walking and hunger-cawing, for grain-sight-pecking, grain-flirting, grain-sight-cooing, grain-sight-walking, hunger-ground-scratching, and for throwing objects out of the beak, in the sub-cerebral brain; and all the corresponding arcs, except perhaps the first two and the last, appear to suffer increased injury by removal of a thin slice from the top of the brain stem.

12. The decerebrated pigeon is sometimes attracted by water and sometimes by grain.

13. It is sometimes repelled by the approach of water, sometimes by that of grain.

14. There are inhibitory mechanisms in the sub-cerebral brain.

15. The decerebrate pigeon does not react to the same stimuli always in the same way, i.e., its reactions are not fatal like those of an automaton.

16. The decerebrate pigeon is not indifferent to its surroundings.

17. After cerebral ablation it takes the pigeon many months to show all the properties of the residual brain, and therefore it seems reasonable to infer that the connections of some of the sub-cerebral arcs are not *normally* wholly patent; which implies that the corresponding functions are, in the pigeon's present stage of brain evolution, *normally* mediated chiefly through the cerebrum.

18. There is no reason to infer that sub-cerebral correlation centers "take over" cerebral functions after cerebral ablation. One may more safely infer that certain ancient sub-cerebral connections, which had become less patent because of, or by means of, the production later of more patent connections in alternative or shunt arcs in the cerebrum, again become sufficiently patent for reaction after removal of the shunt arcs with the cerebrum.

An attempt is made to show how use and disuse may be potent factors in determining the location of brain centers.

Certain laws of brain evolution and of education, suggested by the findings in the light of present day knowledge, are tentatively formulated.

The findings are made the basis of a new statement of the functions of the cerebrum and of the sub-cerebral brain.

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STUDIES IN BLOOD PRESSURE ESTIMATION BY INDIRECT METHODS

III. The Movements in the Artery under Compression during Blood Pressure Determinations

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In the efforts that have heretofore been made to ascertain the mechanism and the significance of the signs commonly employed in the determination of the arterial pressures in man, the main methods of investigation have consisted in observing with the unaided eye the character of the movements of the artery in the compression chamber or in recording the volume or the pressure oscillations that are produced in the compression chamber by the pulsatile movements of the compressed artery as a whole, and comparing these with the arterial pressures. The inadequacy of simple inspection of a series of movements, some of whose phases run their course in a small fraction of a second, as a means of ascertaining the character of that motion and its significance is so obvious as to make comment unnecessary. But that a record of the movements of, or produced by, a compressed arterial segment *as a whole* in many ways also is inadequate may not be so apparent. The inadequacy of the latter method is due to the fact that the pulsatile changes occurring under the compressing band, or in the compression chamber, are effected by processes that are propagated as waves along the artery, so that at any given instant every point along the course of the compressed segment is in a different phase of the propagated disturbance; the record taken from the whole of the artery is a composite of a number of different processes occurring at the same time.

Ever since we first became interested in this problem (1) we have realized these shortcomings of the methods available for the study of the movements of a compressed artery and consequently have come back to the problem whenever new methods of attack seemed to hold out the hope of arriving at a more definite understanding of the under-

lying phenomena (2), (3). The present paper is based upon the use of methods by which records can be made of the movement of points along the course of an artery *in situ* while and where it is being subjected to air compression (4). In certain ways the records obtained by these methods, even with the aid of collateral evidence (5), are not easy to interpret, but they have, we believe, shed much new light upon the movements of the compressed artery and upon the mechanism and significance of certain of the blood pressure signs and have helped materially toward clearing up certain of the moot phases of the blood pressure problem.

METHODS

For the purpose of recording the movements of points on the compressed segment of artery two different types of compression chambers have been employed. With each the recording methods were different.

Chamber for records by reflected light. This chamber (fig. 1) was made in three parts, namely a bottom, a middle piece and a cover. The bottom, *a*, was made of brass and had the form of a rectangular tray with a depth of about 6 mm. The middle piece, *b*, was an oblong rectangle of the same material and dimensions as the tray, except that it was about 17 mm. deep. It fitted accurately the rim of the tray-like bottom. Into the contiguous rims of these two parts were sunk half-tubes, *d*, in such a way that when the parts were brought together the half-tubes formed complete tubular outlets through the end walls of the chamber. These tubes, which provided the passageway for the artery, *e*, through the walls of the chamber, measured 13 mm. in length and had a bore of 5 mm. They extended about 4 mm. beyond the outer surface of the chamber and about 5.5 mm. beyond the inner surface. Of the ends of the half-round tubes projecting into the box all was cut away excepting thin strips where they came into apposition when the bottom and middle pieces were joined. These strips, broadened into shelves, *f*, served to clamp together and support the leaflets of the valves which were for the purpose of blocking the exit of air alongside the artery. The valves were put together as follows: A piece of soft, wet, peritoneal membrane measuring about 2.5 by 2.0 cm. was laid upon the lower half-round part of the tube so that the inner edge of the membrane reached just to the inner end of the supports, *f*, mentioned above, while the outer edge projected a considerable distance beyond the outer projection, *d*. The bottom of the chamber was then slipped under the artery until the latter lay in the half-round

grooves and over the peritoneal membranes. Then two similar pieces of peritoneal membrane were exactly superimposed upon the lower pieces but with the artery intervening, and these pieces also were closely applied to the surface of the upper trough. The middle piece of the chamber was then fastened in place. Now the outer projecting ends of the valve flaps were reflected over the end of the projecting

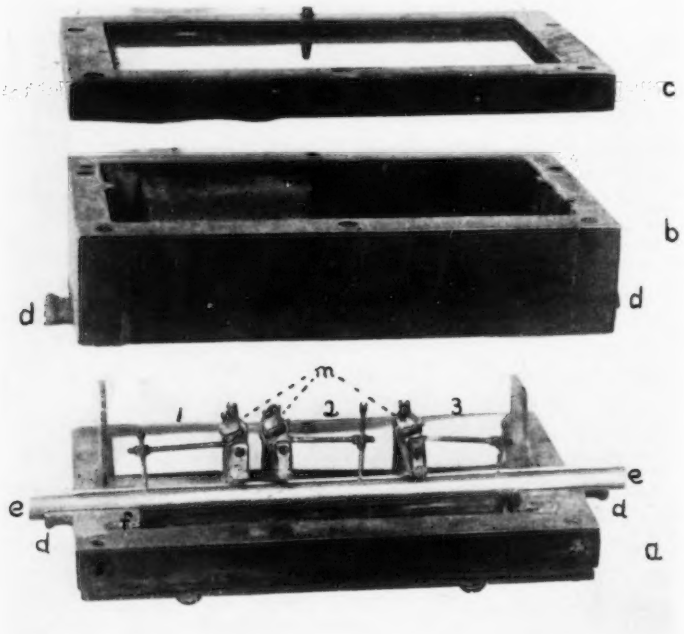


Fig. 1. Mirror lever compression chamber, natural size. Description in text.

tube and so tied to it that an air-tight joint was made between the metal and the peritoneal membrane. Then the top, *c*, consisting of a plate of glass set in a metal rim, was screwed down to the box. Rubber gaskets served to make the joints of the chamber air-tight. When air was forced into the chamber through the wide tubulation provided for the purpose, the inner free edges of the wet peritoneal membranes, acting as valve flaps, closed down against the artery and so tended to

prevent the escape of air alongside the artery. Not infrequently, however, this joint was somewhat incompetent, owing probably to the folds into which the peritoneal membrane was thrown. In order to maintain the desired air pressure in the chamber it was necessary in such instances to keep the chamber connected with a large reservoir containing air under the desired pressure and saturated with water vapor. The length of the chamber from outer end of tubulation to outer end of tubulation was 82 mm.

The movements of the artery within the chamber were followed by means of small, horizontal levers. These levers, three in number (*1*, *2* and *3* in fig. 1) were so mounted on posts erected on the floor of the chamber that all moved in one plane, one perpendicular to the floor; and the posts were so constructed that the artery could be brought directly under and parallel to the levers. The levers were held to the artery by gravity. The contact between the artery and the lever was made by means of a long, light watch-screw threaded vertically through the free end of the lever. The pivoted end of each lever carried a minute, approximately horizontal mirror, *m*, which could be tilted laterally by means of another watch-screw. By means of these two screws the plane of the mirror could be brought exactly into the desired position relative to an incident beam of light. As the top of the chamber could be removed without disturbing the joint between the bottom and middle pieces, this adjustment of the plane of the mirror could be made between observations and without disarranging the artery.

When it was desired to watch or to record the motion of the light reflected from the mirrors on the levers the artery was first fixed in the instrument as described above. Then the position of the animal was adjusted so that the chamber with its contained artery lay in the horizontal plane, and so that the artery was directly below and parallel to a horizontal screen or the slit of a downwardly directed photoregistering apparatus. A beam of light from an arc lamp was thrown through the glass top of the chamber upon each mirror and each mirror was then adjusted by means of the screws referred to above until its beam was reflected back through the glass top to the desired position, and in focus, on the slit of the camera or on the screen. The paths of the beams of light lay approximately in the plane of motion of the levers. Any deviation from this position that caused recognizable displacement of the pictures on the sensitized paper was allowed for in measuring the records. Condensation of water vapor on the inner surface of the glass cover, with the consequent dispersion of light, was obviated by applying heat to the cover.

The first lever, *1*, and the second lever, *2*, rested on the artery 15.7 and 37.3 mm., respectively, below the outer end of the proximal (with respect to the heart) orifice of the chamber, and the third lever, *3*, 67.2 mm. below that orifice and 15.3 mm. above the outer end of the distal orifice. The distance between the levers and the photoregistering apparatus always was such that elevation of the free end of the lever through 1.23 mm. caused the beam of light to move over the sensitized paper through, 30, 33 and 28 mm. for levers *1*, *2* and *3*, respectively. Roughly, therefore, 1.0 mm. of deflection on the record denotes a movement of the arterial wall through 0.04 mm.

The inherent vibration rate of the levers was determined by recording the vibrations set up when they were allowed to fall upon a stretched sheet of rubber dam. The time required for the completion of a double vibration diminished as the amplitude of the vibrations thus called forth died away. With the values thus determined a curve was constructed from which the time required for the completion of a fling of any amplitude could be estimated. In order to indicate the capabilities of the levers, it may be stated, by way of example, that roughly, double vibrations of a total amplitude of 25 mm. were completed in 0.03 second; of 10 mm. amplitude in 0.015 second; and of 5 mm. amplitude in 0.01 second. Lever *2* was somewhat more sluggish than the others. Levers *1* and *3* behaved so nearly alike that time differences indicated by them on the records may be regarded as absolutely correct. The pulses written by levers *1* and *2* are inverted; those written by lever *3* are erect.

As the distal end of the mirror lever rested on the highest point on the circumference of the artery when full, it follows that an ideal record would be given only by an artery that flattened in the plane perpendicular to the plane of motion of the lever, and one that suffered no translateral motion as a result of the action on it of the pulse or of the compression. Only under these circumstances would the motion of the lever measure accurately fluctuations in the fullness of the artery. But for many reasons these ideal conditions were difficult of attainment.

1. In the first place it was not always possible to so arrange the artery in the compression chamber that it would flatten under compression in the plane perpendicular to that in which the levers moved. To be sure, the valves at the orifices worked in this plane and therefore imparted to the artery a tendency to flatten horizontally. But despite this directive influence, and even despite directive manipulation of the artery, it was impossible at times to get it to flatten exactly in the

horizontal plane. Indeed, the femoral artery as prepared in the animal experiments cannot, on account of the presence of ligated branches, be brought accurately into one plane under compression. Naturally, only those experiments were suited to our purposes in which the artery did flatten essentially in the horizontal plane. On this account the carotid, in some respects, would have been better than the femoral artery, and it was employed in some of the special experiments. But in other respects the carotid is not quite as satisfactory as the femoral. Thus occlusion and deocclusion of the carotid effects wider consequent fluctuations in the general arterial pressure. Furthermore, when the carotid artery is occluded the pressure beyond does not fall to the same degree as it does in the case of the femoral; the conditions in the latter case, therefore, simulate somewhat more closely those obtaining while determining the arterial pressure in man.

2. The flattened artery sometimes exhibited with each pulse slight rotational movements around its axis. When this occurred the movements of levers of course did not indicate changes in arterial bore alone.

3. Translational movements of the artery or movements having the effect of translational movements might be caused in a number of ways:

a. The changing weight of blood in the artery might alter the degree to which it sags. To determine whether this might be a factor, with the chamber arranged as for making a record but with the cover removed, determinations were made of the motion of the lever caused by adding weights to its free end. It was found that a weight of 0.02 gm. caused the reflected beam of light, with the magnification used in the animal experiments, to move downward 0.7 to 1.0 mm. As will be seen, the amount of blood held in the artery during diastole certainly does not exceed 0.2 gm. in weight, at least not until decompression has proceeded almost to the diastolic level. Furthermore the weight, when added to the lever, is distributed over a very much more limited area of the artery than is that of any blood the artery might contain, and therefore must cause far more motion of the lever than would the entrance into the artery of an equal weight of blood. It may be concluded, therefore, that though gravitational movements of the artery due merely to variation in its blood content may occur, they must be insignificant unless, perchance, they are magnified by other circumstances.

b. As is well known, distention of an artery causes it to stretch not alone radially but also longitudinally. The direction in which longitudinal stretching would cause the axis of the artery to move would depend upon circumstances which are too obvious to require enumera-

tion; of these the directive action of gravity would always be present. It therefore is to be presumed that as the pressure in the artery rises with systole or with decompression, motion of the artery downward would occur more commonly than motion in any other direction.

c. The orifices of the compression chamber were made no larger than was necessary to just allow an artery of average size a clear passage through them. This was necessary in order to insure a fair degree of competence of the valve joints. While adjusting the artery in the chamber every effort was made to center it in the orifices. This, however, was not always possible. As it was necessary to make room for the compression chamber under the artery and as, in addition, gravity tends to move the artery downwards, it is justifiable to infer that such accidental contacts as may have occurred were more frequent at the lower than at the upper edge of the orifices of the chamber. The effect of such accidental contacts would be to move the artery en masse away from them as it filled.

Chamber for records by shadow registration. In an effort to obtain a basis for the evaluation of the defects inherent in the registration of caliber changes of the artery by means of the levers of the compression chamber described above, a chamber was devised by means of which the shadow cast by the artery flattening horizontally in the path of a horizontal beam of light could be recorded photographically.

The construction of this chamber differed from that of the preceding one mainly in that the lateral walls, instead of the top, consisted of glass. As it was necessary to pass the light horizontally through the chamber at the level of the artery, the joint between the bottom and middle piece of the chamber had to be avoided; and yet in order to get the benefit of the directive influence of horizontal valve leaflets on the flattening of the artery, it was necessary to provide a means of adjusting the leaflets horizontally. This was accomplished by making end walls that slid into place from above in grooves. These ends carried the upper half of the tubular openings for the passage of the artery, and the upper supports for the valves. After the artery and valve leaflets had been properly adjusted these ends were shoved into place and the box was then closed by tying back the leaflets and by screwing down the lid. Through this arrangement the movements of the upper and lower walls of the artery in any selected circumference could be recorded. These movements were magnified 11.2 times, and inverted, on the record.

The advantage this method has over the mirror lever method is that translational movements of the artery in a vertical direction can be recognized and allowed for. But the method is fraught with other possible sources of error. The main ones are 1, that if the artery should fail to flatten exactly in the horizontal plane its image while flat would give a false impression of the thickness of the artery in the completely, and to a variable degree in the partially, collapsed state. 2, Torsional movements of the flattened artery, if present, would give the impression of changes in caliber. And 3, the artery when flattened beyond a certain degree, as has long been known (6), (7), (1), takes on the form of an hour-glass on section; on this account the diastolic changes in caliber occurring in the mid-line during that degree of flattening are not shown; and for the same reason it is not to be expected that under the higher compressing pressures the pre-systolic movements will be as clearly brought out as by the mirror levers, which always record the movements of the mid-line.

The Korotkoff sounds were followed by a stethoscope (3) which surrounded the artery just below the compression chamber. It should be recalled in this connection that in the dog sounds having the characteristics of the second phase often are lacking, and that the artery emits sounds even when it is under no compression whatever; that is to say, there is no phase of silence or so-called fifth phase. Otherwise, however, the Korotkoff sounds in the dog are identical with those heard in man. Therefore, in order to avoid confusion, the designation of the phases in general use has been employed. Before the fourth phase begins the third-phase sounds, which may be very intense, usually diminish slightly in intensity and in almost imperceptible stages. This is usually the case in man also (8). The fourth phase is indicated by a change in the character of the sounds from loud and clear to dull and muffled. In the dog this change usually is quite sharp and unmistakable, and after it there is no other sharp change in the quality or loudness of the sounds, but only a gradual diminution in intensity. The beginning of the first and of the fourth sound-phases was signalled on the record. There was obtained in this way an absolutely unbaised means of comparing the sound-phases with the events on the record. It should be added that in some of the experiments the noise made by air escaping alongside the artery made it very difficult, even sometimes impossible, to accurately signal these points.

On all of the records the time was recorded in fifths of seconds. The paper moved quite uniformly and, in the case of the mirror records,

usually at the rate of about 80 mm. per second. Sharp changes in the direction of motion of the beams over the record could be read with the aid of a binocular magnifier through a glass coördinate system to within 0.2 mm., and therefore with certainty to within 0.003 second.

In none of the animal experiments was a record made of the pulsatile oscillations of pressure within the compression chamber. It has been stated that on account of leakage of air through the peritoneal membrane valves guarding the orifices of the compressed chamber it was necessary to keep the chamber in connection with a large reservoir of air. The presence of this large air chamber practically eliminated the pressure oscillations usually produced by the pulse. Satisfactory oscillation records could have been obtained in only the one experiment of the series in which the valves were completely competent. Many records have, however, been accumulated of compression oscillations made in other connections. So constant has been the relation of the oscillation phases to the sound-phases in these records that it seems quite justifiable to assume that in the present experiments the oscillation records, had they been made, would have had the usual configuration and relations.

Neither was the compressing pressure directly recorded. But when steady pressures were employed the manometer was read by an assistant; and when the records were made while the compressing pressure was falling either the rate of fall, from before the time the pulse began to penetrate and until some time after the beginning of the fourth sound-phase, was kept fairly constant, and before and after making each record the compressing pressures corresponding with the beginning of the first and of the fourth sound-phases were determined; or an assistant signalled on the record each 10 mm. Hg. decrement of the compressing pressure. In the former case, deductions dealing with the relation between compressing pressure and caliber of artery, for instance, can be regarded as only rough approximations, and the curves we have made by plotting caliber of artery against compressing pressure as only approximately correct. In the latter case it is possible to determine fairly accurately, by means of interpolation, the compressing pressure under which each pulse was recorded; the plotted curves therefore represent relations correctly.

The animal employed was the dog. Morphine and ether were administered in the usual way. The operative procedures for the application of the lever chamber consisted merely in cannulation of the trachea and preparation of the ileo-femoral artery for its insertion into the com-

pressing chamber and the artery stethoscope, by tying all of its lateral branches. The shadow compression chamber was applied to the carotid artery because it needed more room than the lever compression chamber, and a clear way for the horizontal beam of light. This was obtained by cutting away the trachea after inserting a cannula into it as low down as possible, and then removing the sternohyoid and sternothyroid muscles of both sides, the tip of the transverse process of the 6th cervical vertebra and the tip of the manubrium sterni.

A number of schemata were devised in an effort to arrive at an understanding of certain of the phenomena brought to light through study of the behavior of the artery compressed *in situ*. It is convenient to defer the description of these schemata until the phenomena are considered for whose elucidation they were planned.

RESULTS

The mirror lever compression chamber has been successfully applied to six animals. Careful qualitative observations were made in each case, and from three of the animals, which will be designated dogs III, IV and V, a number of complete records of the movements of the arterial walls during decompression were obtained for quantitative study.

Dog III was a large animal whose femoral artery was not, however, particularly large. The Korotkoff sounds were of fair intensity (there was no second phase) but owing to the noise made by air escaping from the compression chamber through the joints of the valves the experimental error of signalling the first- and fourth-phase sounds was, we believe, unusually large. Levers *1* and *2* did not record clearly. At critical points the records were, however, clear enough for purposes of study. The artery seemed to flatten nicely in the horizontal plane. The pulse rate was rather rapid, 156 per minute. The number of pulses during the first-to-fourth sound-phase was forty-seven. It is therefore assumed that the compressing pressure fell at the rate of 1.5 mm. Hg. per pulse.

Dog IV was a large bulldog with a fine large artery, loud Korotkoff sounds (without a second phase), and a relatively slow pulse, 110 per minute. The part of the artery under lever *1* flattened in a decidedly oblique plane; under the other levers, however, the artery flattened nearly in the horizontal plane. The lower end of the artery was considerably smaller than the upper. There was very little escape of air

through the valves and therefore the sound-phases could be sharply signalled. While making record 1, the one which is reproduced as figure 3, the note was made, before the record was developed, that the first sound was signalled about 5 pulses late. These pulses have therefore been included in the first-to-fourth sound-phase period. On this basis the number of pulses in this period was 39. The first and fourth sound-phases began at compressing pressures of 138 and 78 mm. Hg., respectively. The pressure decreased approximately at the rate of 1.5 mm. Hg. per pulse.

Dog V was also a large bulldog with a large artery. The valves of the compression chamber did not leak at all; therefore the sounds, which were quite characteristic and loud, but again without a second phase, could be clearly heard; the phases were signalled with perfect certainty. The pulse rate was rapid, 150 per minute. The rate of decompression was signalled on the record. The first and fourth sound-phases began, as indicated on the record, under compressions of 182 and 122 mm. Hg., respectively. The compressing pressure fell somewhat more slowly during the first part of the first-to-fourth sound-phase period; here the rate of decompression may be taken as 1.0 mm. Hg. per pulse. During the rest of this period the rate of decompression was about 1.33 mm. Hg. per pulse.

The shadow compression chamber was successfully applied to three animals, and a number of photographic records was obtained from each of them. They will be designated as dogs VI, VII and VIII. The fall in compressing pressure was signalled on the record in steps of 10 mm. Hg. Though the attempt was made, the sound-phases could not be very successfully signalled in these experiments owing to the noise of escaping air through the valve joints.

Dog VI. In this case the artery when flattened was not exactly in the horizontal plane, at least in the distal part of the chamber; as a consequence the motion at the distal end with the advent of the pulse was in part a rotation of the plane on the axis of the artery. The records were taken from the artery 5.5 cm. distal to the outer end of the proximal orifice of the chamber.

Dog VII. This artery flattened in the shape of a crescent opening upwards, due apparently to partial drying of the upper surface of the artery. The ends of the crescent were in the horizontal plane. The first sound-phase began very gradually and it was therefore difficult to accurately signal its beginning. The fourth sound-phase seemed to be satisfactorily signalled. The photographed circumference of the artery was located 6.2 cm. distal to the proximal orifice of the chamber.

Dog VIII. In this case the artery flattened beautifully in the horizontal plane. The noise of the escaping air made it impossible to signal the sound-phases. The slit exposing the artery to the camera was located 5.7 cm. beyond the proximal orifice of the chamber.

It is convenient for purposes of referring to the records to use a system of numbering the pulses which will in itself be somewhat descriptive. With this end in view the first pulse, during decompression, that produces in the tracing made by lever *3* a clear anacrotic limb is designated *pulse 1*. This lever is close to the lower end of the compression chamber and presumably is quite close to the beginning of the lower conical closure of the artery. It may therefore be assumed that when the pulse reaches lever *3* it practically has succeeded in completely penetrating the compressed artery. The subsequent pulses are numbered consecutively to the end of the record; while the pulses preceding *pulse 1* also are numbered consecutively but in the direction contrary to the motion of the paper; and, to distinguish them, they are prefaced by the minus (−) sign.

Successive records obtained from one and the same animal are usually strikingly similar, though from time to time unessential variations may be seen. But records obtained from different animals in certain respects differ very markedly in appearance from one another. These differences undoubtedly are to be attributed in large part to the technical difficulties to which reference has been made above. To have increased the number of animals would merely have resulted in multiplying the variety of records. The records are full of detail, every feature of which, we feel convinced, is of some significance with respect to the processes enacted in the compressed segment. But many of these details, owing in part to differences in different animals, have thus far failed of interpretation. It seems justifiable to maintain, however, that if the signs, ausculatory, oscillatory, etc., etc., employed for the purpose of determining the arterial pressures by indirect methods are really indicative of the systolic or diastolic pressures, as the case may be, it should be possible to find in the records movements capable of accounting for the signs, and so delimited as to show that they indicate the pressures they are supposed to indicate. It will become clear that this can be done. The present paper concerns itself only with those features of the records that seem to be of significance with respect to the blood pressure problem.

Preliminary theoretical discussion. Consideration of the records is facilitated by discussion from a theoretical standpoint of the movements

a point on the upper edge of the artery would be expected to make through the action of the forces at work in a decompression experiment. Leaving out of consideration at first all of the factors that tend to impede the motion of the wall imparted by the pulse other than the elasticity of the wall itself, we arrive at the reconstruction illustrated by the heavy lines in figure 2. The abscissae of this system represent time in the pulse cycle. Of the two sets of ordinates to the left, one gives the radius of the artery in millimeters, the other, the distending

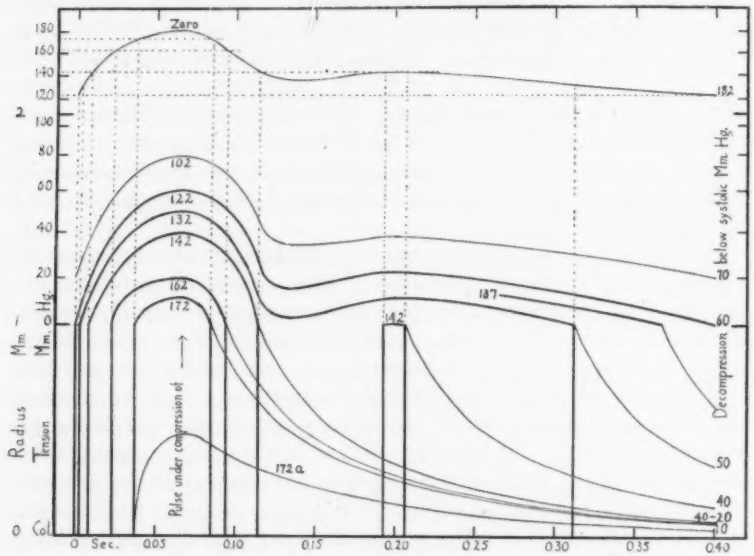


Fig. 2. Diagram constructed to illustrate the theory of emptying of a compressed segment. Description in text.

pressures that impart to the artery the radii indicated by the former set. The first step from *Col* (collapse) to 1 mm. (and 0 mm. Hg.) represents the increase in radius that takes place with the arrival of the pulse at a time when the compressing pressure is higher than the diastolic, but lower than the systolic pressure. The wall of the artery here moves at once from the position of complete collapse to its *just round*[1] or zero

[1] We prefer the term "just round" to the one, namely, "zero position," employed by MacWilliam and Melvin (9) to designate the size of the round but unstretched artery.

size, and then higher, if stretched. The magnitude of this step to the "just round" size has been chosen arbitrarily. The size of the steps, under *Tension*, above this step have been determined by calculation from MacWilliam's experiments on the relation of cubic capacity of a relaxed artery to internal pressure (10). Into this system of coördinates we have fitted a typical pulse (designated by *zero*) of dog V. This pulse has been drawn in approximately the same form and time relations as the pulse obtained from this animal by the mirror levers under complete, or practically complete, decompression; and its amplitude, as given by its systolic and diastolic pressures as determined by experiment, has been made to conform with these coördinates. The character of the motion of the point on the arterial wall under several compressing pressures namely, 122, 132, 142, 162, 172 mm. Hg., all within the systolic-diastolic, or *critical*, range, arrived at by reconstruction, is indicated by the heavy-line curves. The method of determining the dimensions of these pulses is indicated by the dotted lines. The artery rises from or falls to collapse (*Col*), at the instant in the pulse cycle the intra-arterial pressure rises above or falls below the compressing pressure, respectively. And the degree to which the artery fills above the just round size depends upon the difference between the systolic pressure and the compressing pressure in relation to the extensibility of the arterial wall. The effect that counterbalancing a part of the internal (arterial) pressure with a pressure from without that is less than the diastolic pressure will have upon the amplitude of motion of the point also is indicated by the diagram (compare pulses *102* and *zero*).

The influence upon the motion of the point exerted by some of the factors that modify the collapse (or filling) of the artery may now be considered. One of these factors is the resistance to outflow from the compressed segment as determined by the resistance within the segment itself. The rate of outflow, according to Poiseuille's law, will vary as the fourth power of the radius. It is obvious, therefore, that it will slow down very rapidly as the lumen narrows. This effect, together with that of inertia, which is another of the modifying factors, will tend to delay the emptying of the artery, so that instead of taking place instantaneously when the interarterial pressure falls below the compressing pressure, it will take place somewhat as is indicated in the figure by the light lines that continue the heavy lines.

But the compressed segment can empty itself only by overcoming the resistance encountered in the parts of the artery contiguous to the

compressed segment. Centrally, this resistance will be determined very largely by the central arterial pressure itself, which here never will fall below the diastolic level. Peripherally, the resistance, from back pressure, at least, at first will be quite low; the main obstruction here during the early stages of decompression will consist of friction. These two resistances, central and peripheral, will merely have the effect of delaying still further the complete pulsatile collapse of the compressed segment, and usually more at the central than at the peripheral end. The effect at the central end will tend to impart to the point a motion having the pattern of the arterial pulse; this effect it is difficult to represent.

Just as inertia and friction tend to delay emptying of the artery so these factors tend to delay filling; actually, therefore, the filling of the artery by the pulse in each stage of decompression will fall short of the theoretical expectation. The way in which we would expect pulse *172*, for example, to be modified by these factors is indicated in the graph by pulse *172 a*.

The graph indicates that just as long as through the operation of such factors as inertia and friction the wall of the artery fails to reach the support it has in complete or almost complete collapse by virtue of contact with the opposite wall or of the high resistance to emptying, and the support it has at or beyond the just round size, by virtue of the elastic resistance of the wall, comparatively slight changes in conditions will produce relatively wide movements of the point on the artery and of the level from or to which the pulsatile oscillations reach.

DIASTOLIC PHENOMENA

The diastolic level or trough line. Turning now to a consideration of the records, it is convenient to deal first with certain of the diastolic phenomena. When a high compressing pressure is applied to the artery the pulsatile movements of the levers practically or actually cease. From the continuation of the line the levers then write, the pulse, when it returns during decompression, ascends. This line and its continuation upon which the pulses are built we designate the diastolic level or *trough line.*

Mirror levers. 1. First depression of the trough line. When the artery is under a compression that completely blocks the pulse the trough lines as defined above invariably slowly sink (see figs. 3, 4, 5, 6 and 7). This sinking occurs whether the compressing pressure is

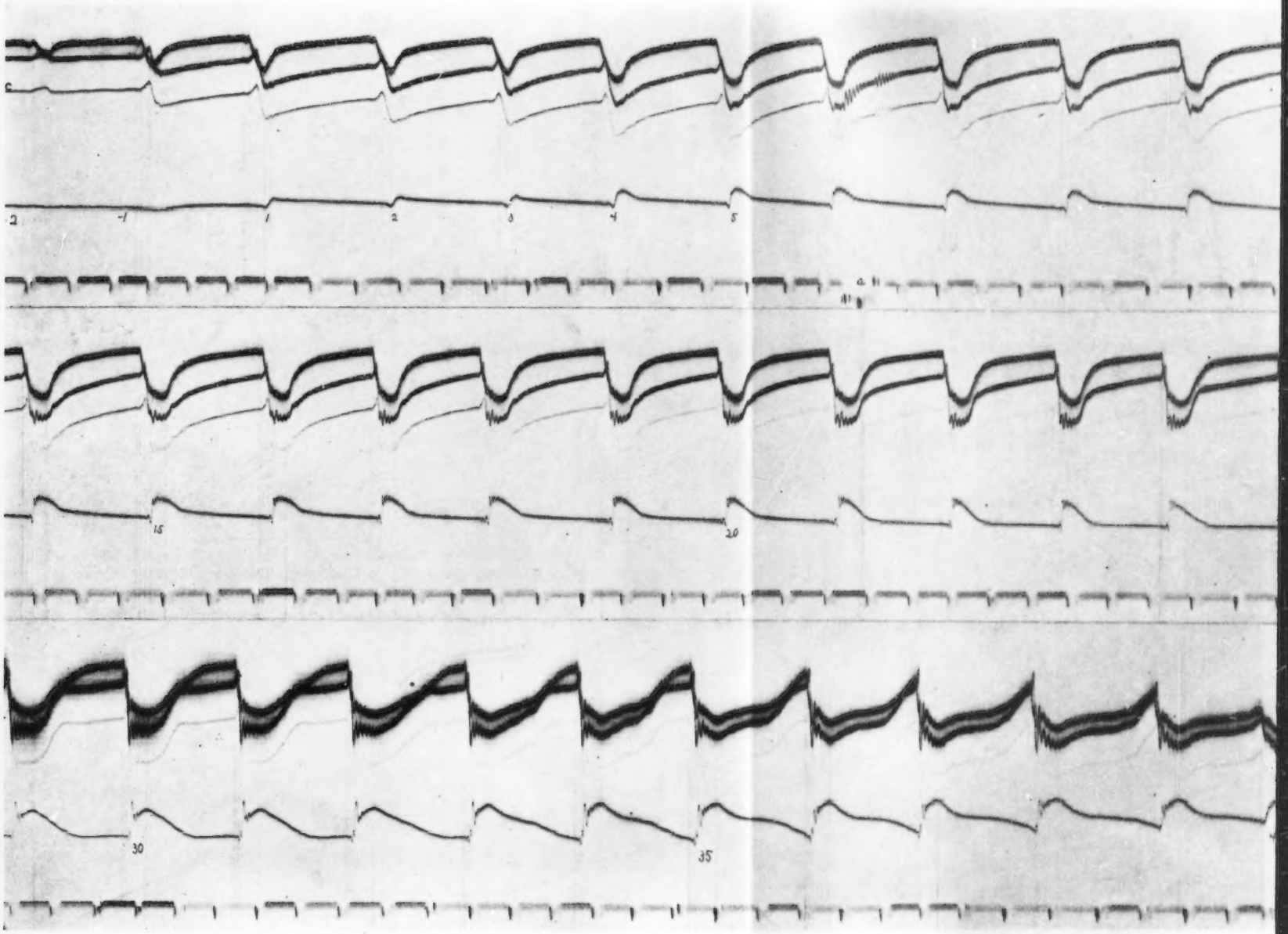
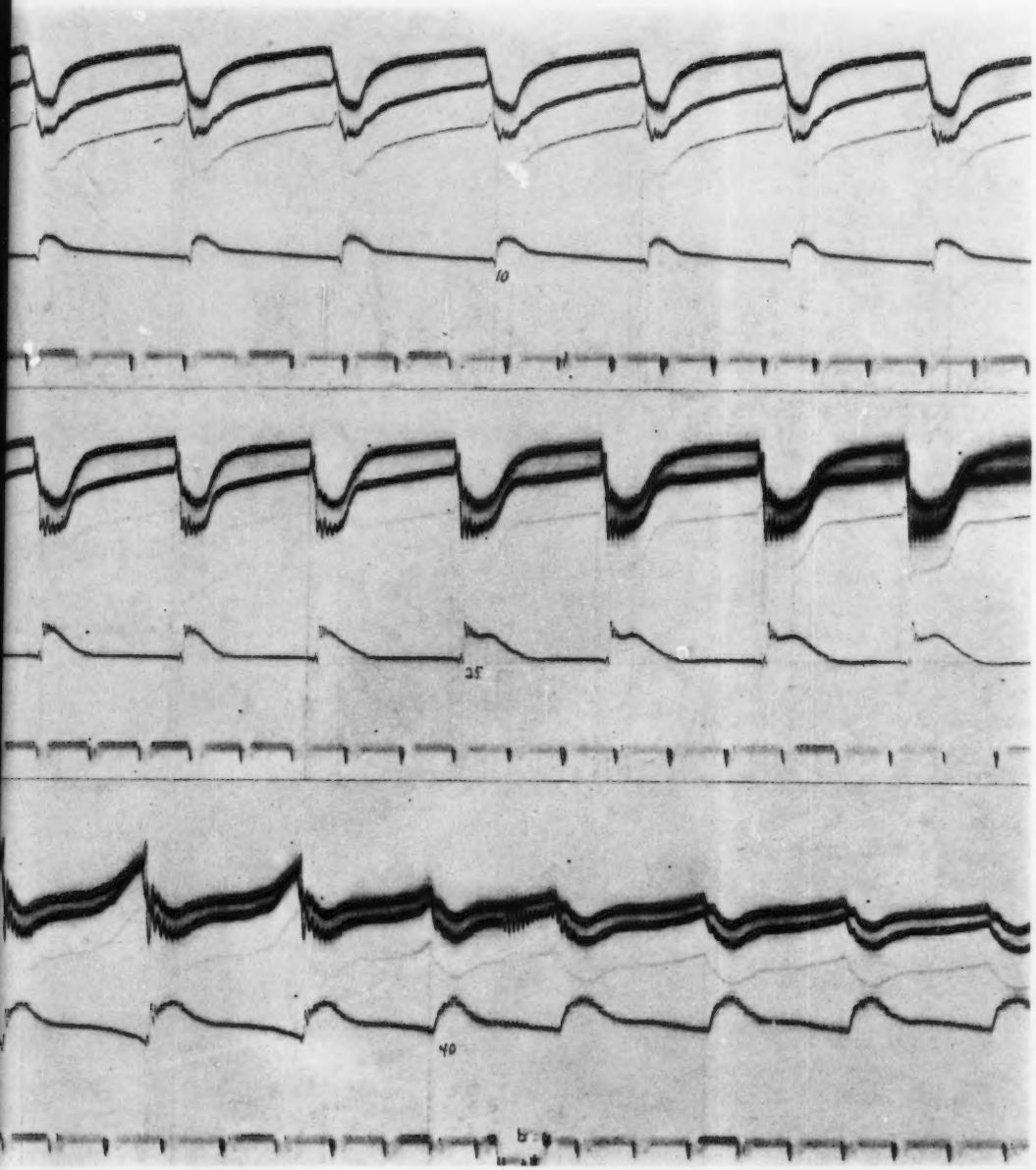
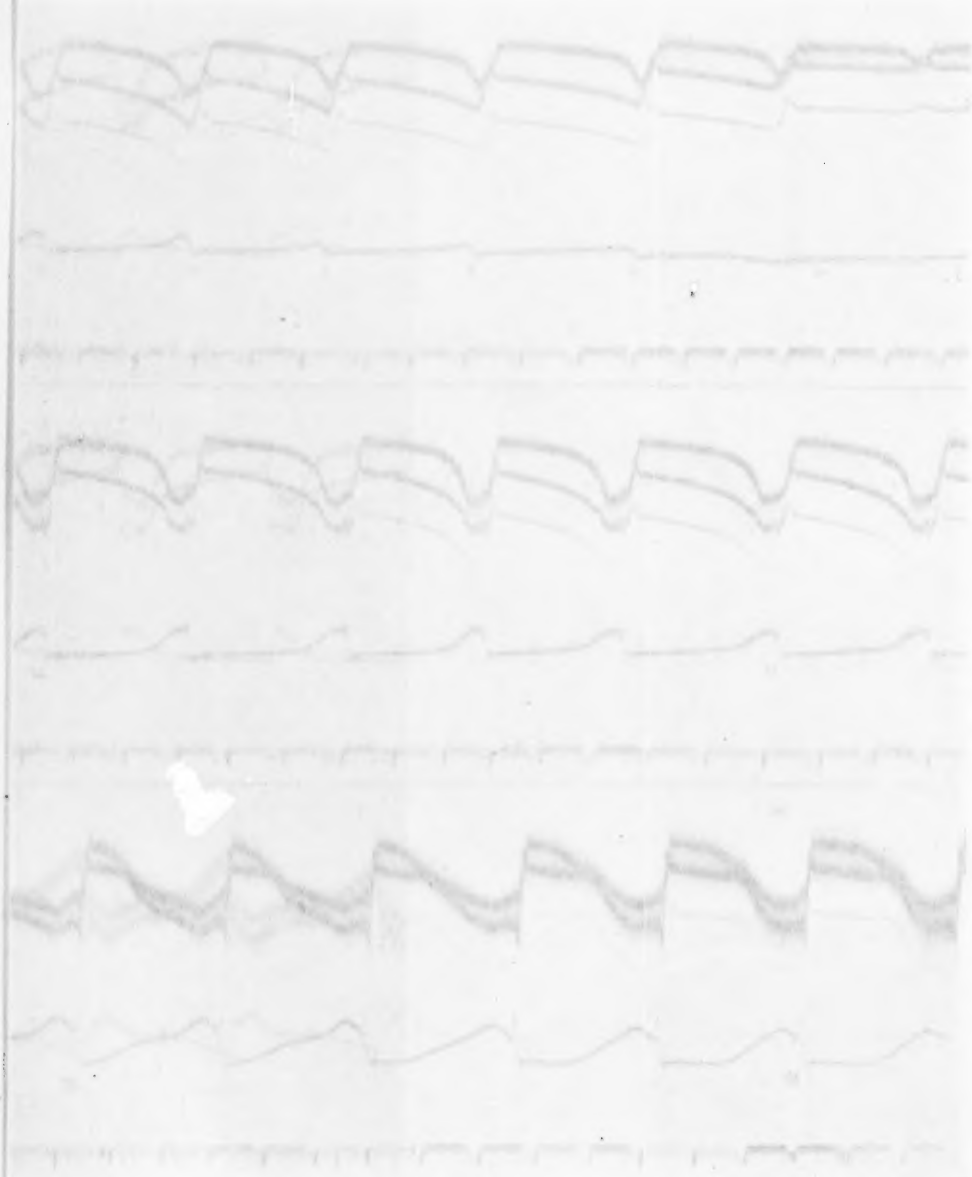


Fig. 3. A complete mirror lever record obtained from dog IV. Reduced to half size. *a* is the signal indicating the beginning of the first phase; *b*, of fourth phase; *c* is a do
5 pulses late. Time in fifths of seconds.



ing of the first phase; *b*, of fourth phase; *c* is a double reflection from lever *2*. The notes state that *a* was about ths of seconds.



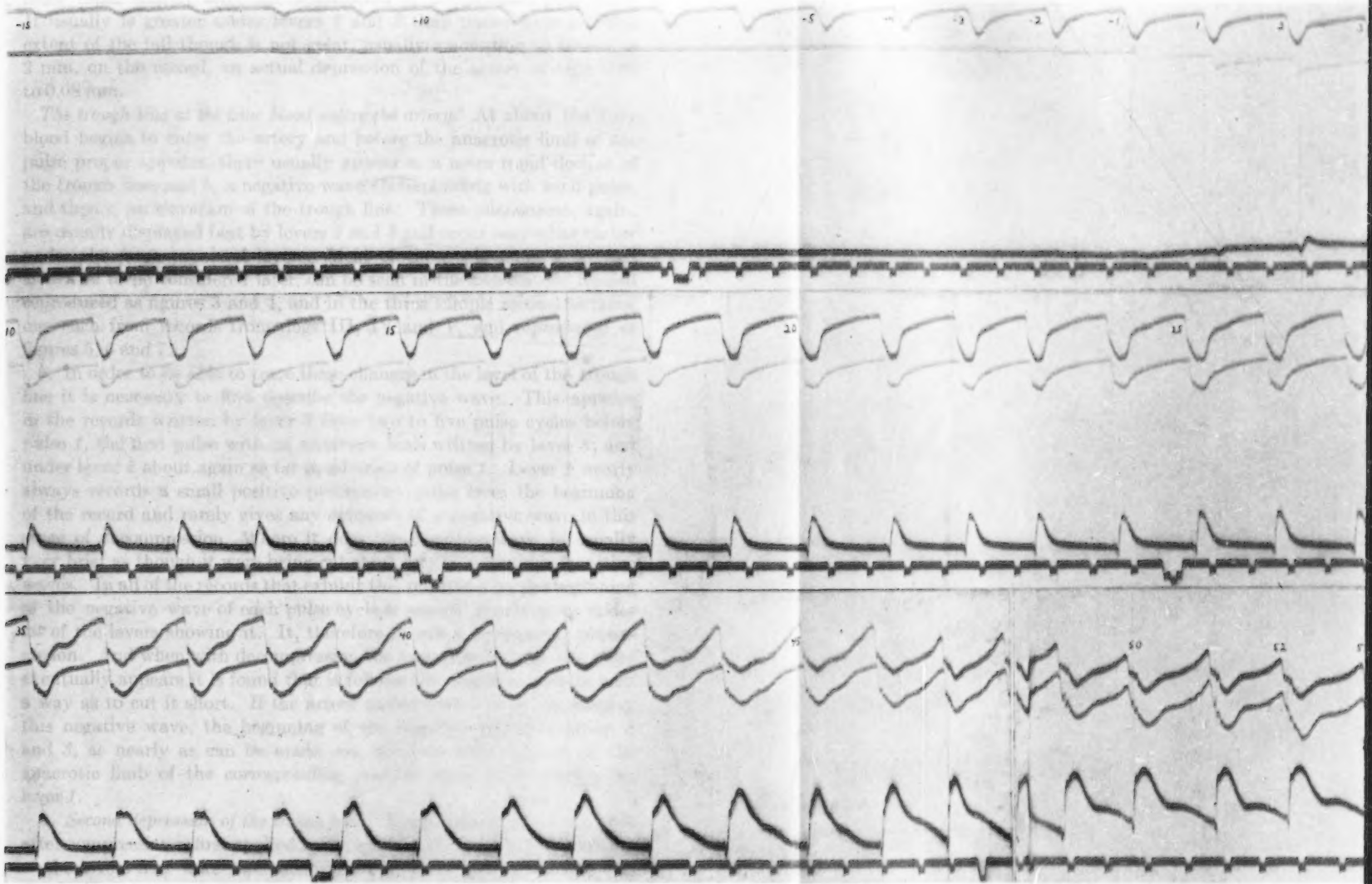
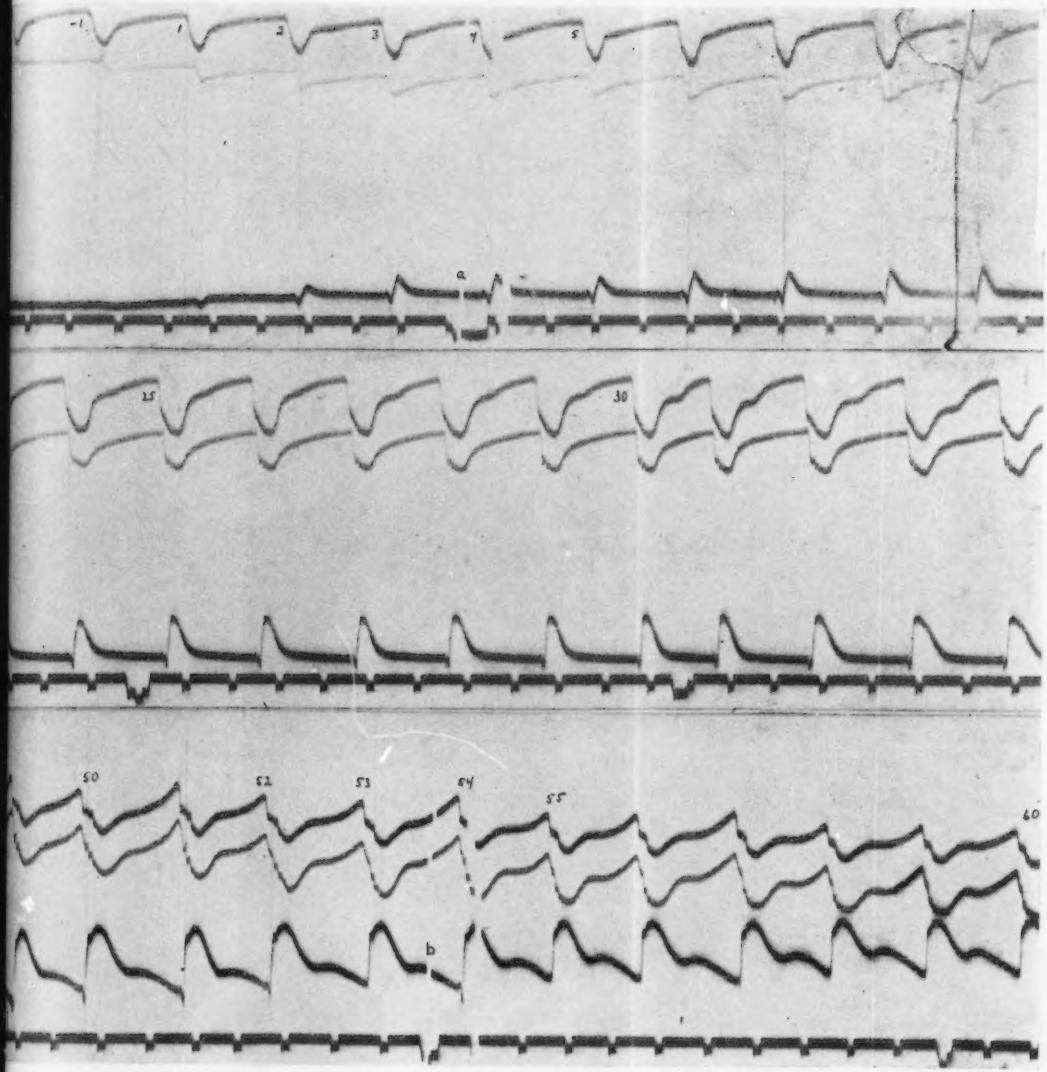


Fig. 4. A complete mirror lever record obtained from dog V. Reduced to half size. *a* is the signal indicating the beginning of the first phase; *b*, of the fourth phase. indicated by the momentary depression of the time record



e first phase; *b*, of the fourth phase. Time in fifths of seconds. Each pressure decrement of 10 mm. Hg. is
f the time record

held at a constant level or is permitted slowly to fall, and seems to be more rapid shortly after the application of compression than later. It usually is greater under levers *2* and *3* than under lever *1*. The extent of the fall though is not great, usually amounting to from 1 to 2 mm. on the record, an actual depression of the artery of only 0.04 to 0.08 mm.

The trough line at the time blood enters the artery. At about the time blood begins to enter the artery and before the anacrotic limb of the pulse proper appears, there usually appear *a*, a more rapid decline of the trough line, and *b*, a negative wave corresponding with each pulse, and then *c*, an elevation of the trough line. These phenomena, again, are usually displayed best by levers *2* and *3* and occur somewhat earlier under the more proximal levers. Most of these phenomena, as well as others to be considered later, can be seen in the two sample records reproduced as figures 3 and 4, and in the three sample reconstructions, one each from records from dogs III, IV and V, and reproduced as figures 5, 6 and 7.

b. In order to be able to trace these changes in the level of the trough line it is necessary to first describe the negative wave. This appears in the records written by lever *3* from two to five pulse cycles before pulse *1*, the first pulse with an anacrotic limb written by lever *3*; and under lever *2* about again as far in advance of pulse *1*. Lever *1* nearly always records a small positive preliminary pulse from the beginning of the record and rarely gives any evidence of a negative wave in this stage of decompression. Where it does the negative wave is usually very brief as though it were being cut short by the preliminary positive waves. In all of the records that exhibit this phenomenon the beginning of the negative wave of each pulse cycle is exactly synchronous under all of the levers showing it. It, therefore, is not a propagated phenomenon. And when with decompression the anacrotic limb of the pulse eventually appears it is found that it follows the negative wave in such a way as to cut it short. If the artery under lever *1* does not display this negative wave, the beginning of the negative waves of levers *2* and *3*, as nearly as can be made out, coincide with the foot of the anacrotic limb of the corresponding positive pulse cycle written by lever *1*.

a. Second depression of the trough line. It has been pointed out that after compression is first applied to the artery a slow, continuous sinking of the levers may take place and that as decompression proceeds this depression of the trough line eventually becomes more rapid. This

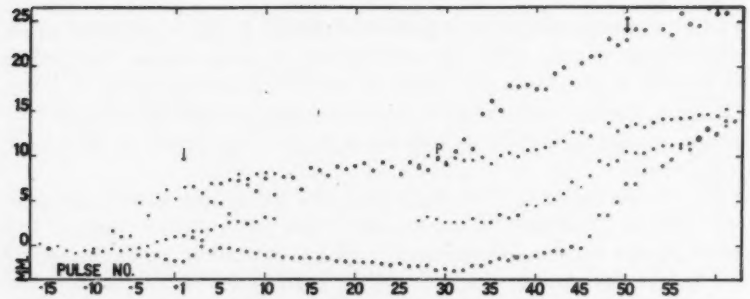


Fig. 5. Diagram of a mirror record obtained from dog III. The level of the record of each of the levers under the highest compressing pressure is taken as the base, or zero, line and the diastolic (lower rows of insignia), and systolic (upper rows of insignia) levels of each pulse are laid off from this line. The pulse numbers are given on the abscissa. The equal spacing between them is based upon the assumption, which is not entirely warranted, that the rate of decompression was uniform. The single arrow indicates pulse *1*. The double arrow indicates the pulse in which the pre-anacrotic phenomenon disappears; disappearance in this case was gradual. *P* indicates the point at which the filling wave appears on the crest of the pulse. Lever *1* is indicated by dots and lever *3* by circles.

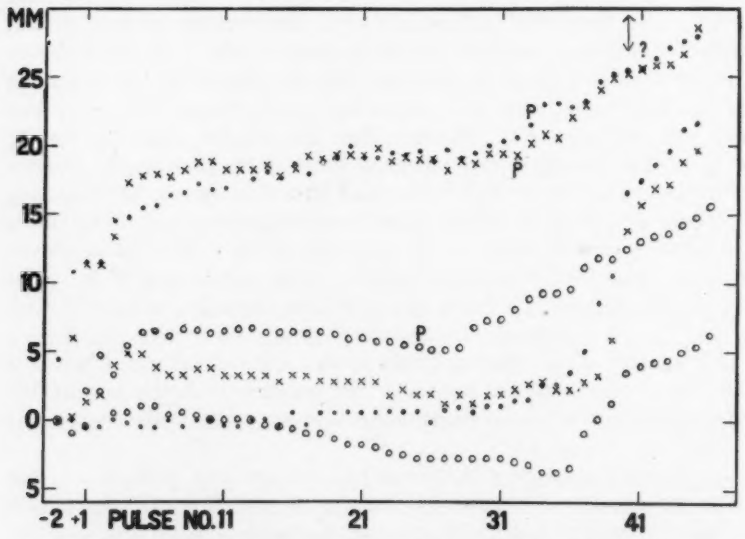


Fig. 6. Diagram of a mirror record obtained from dog IV. Lever *1* is indicated by dots, lever *2* by crosses, and lever *3* by circles; for further description see figure 5.

more rapid, or second, depression, though never very extensive, rarely amounting to more than 1 or 2 mm. on the record, usually is quite distinct. It lasts through only a few pulse cycles, usually not more than from three to five, and it seems to appear earlier in the more proximal parts of the artery. It lasts through a pressure decrement of roughly 5 mm. Hg. Where it attains its best development (again under levers *2* and *3* in all records) it takes place during the pulses that exhibit

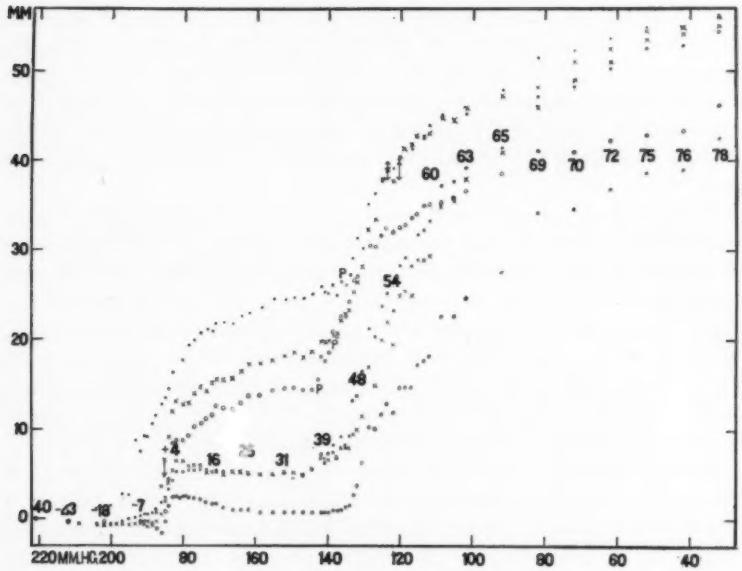


Fig. 7. Diagram of a mirror record obtained from dog V. The numbers of some of the pulses are indicated by the figures in the diagram. The abscissae here are mm. Hg. Lever *1* is indicated by dots, lever *2* by crosses and lever *3* by circles. For further description see figure 5.

the long, low negative wave just described. Indeed, a part of the depression of the trough line is due to the fact that with the first of these negative waves, at least, the ascending phase fails to reach the level from which the descending phase started.

c The second depression of the trough line is followed by a stage in which *the trough line rises*. The changes that bring about this rise differ according as to whether or not the lever is exhibiting the second

depression. If the lever has been writing a pulse without a long, low negative wave and no second trough-line depression, that is to say, if the pulses from the beginning have resembled in form the typical pulse, as is usually true of the record written by lever *1*, the rise results merely through failure of the lever to sink during diastole quite to the level it had at the end of the diastole of the preceding pulse. But, as has been said, in the cycles showing the long, low negative waves the diastolic phase, starting at the close of the descending phase of the negative wave, is either horizontal or only slightly upwardly inclined, the end result being that the trough line either is not materially changed or sinks. Now, however, the lever, during the diastolic phase, actually rises along a gradient that is not inconsiderable, so that the trough line mounts and usually at a relatively rapid rate. Under lever *3* this change invariably begins with, or a pulse or two before, the diastole of pulse *1*.

Within a pulse cycle or two, however, the lever begins to sink during the diastolic phase, so that the cycles begin to take on the appearance of typical pulses. Nevertheless, for some pulses longer diastole continues to terminate on a rising trough line. This rapid rise of the trough line begins earlier under the proximal levers but terminates in all parts of the artery at about the same time. Pulse *1*, as has been seen, always falls in the midst of this rise, and the rise continues for a variable number of pulses after pulse *1*, usually about two or three, occasionally more. The first Korotkoff sound is heard a bit later.

Shadow records. One of the main reasons for making the shadow records was for the purpose of obtaining information with regard to the movements of the trough line that would help in the interpretation of the mirror lever records. It will be observed that whereas similar movements are seen in the shadow records, those that deal with this phase of our problem are not as prominent as they are in the mirror records. We are inclined to believe that this difference is the result of differences in the conditions under which the two sets of experiments had to be done. Amongst these differences the more significant seem to be the more uniform bore of the carotid artery and the relatively slight fall in peripheral pressure when the carotid artery is occluded. Both of these differences would tend to minimize the conditions which, we believe, are in large part responsible for the changes in the level of the trough line under consideration here. One of the shadow records (from dog VI) is here reproduced in full as figure 8. Reconstructions from this record and one each from records from dogs VII and VIII are reproduced as figures 9, 10 and 11, respectively.

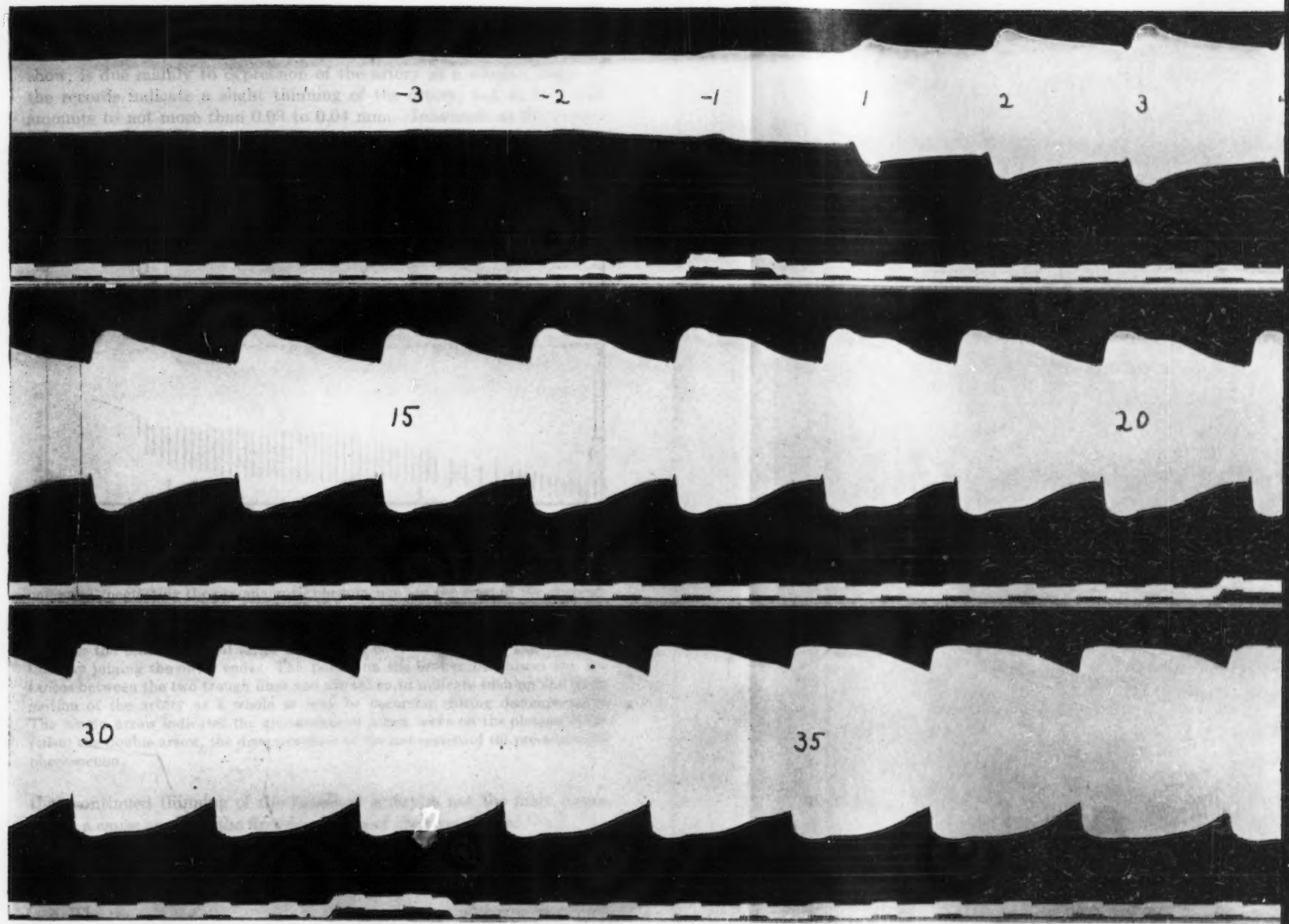
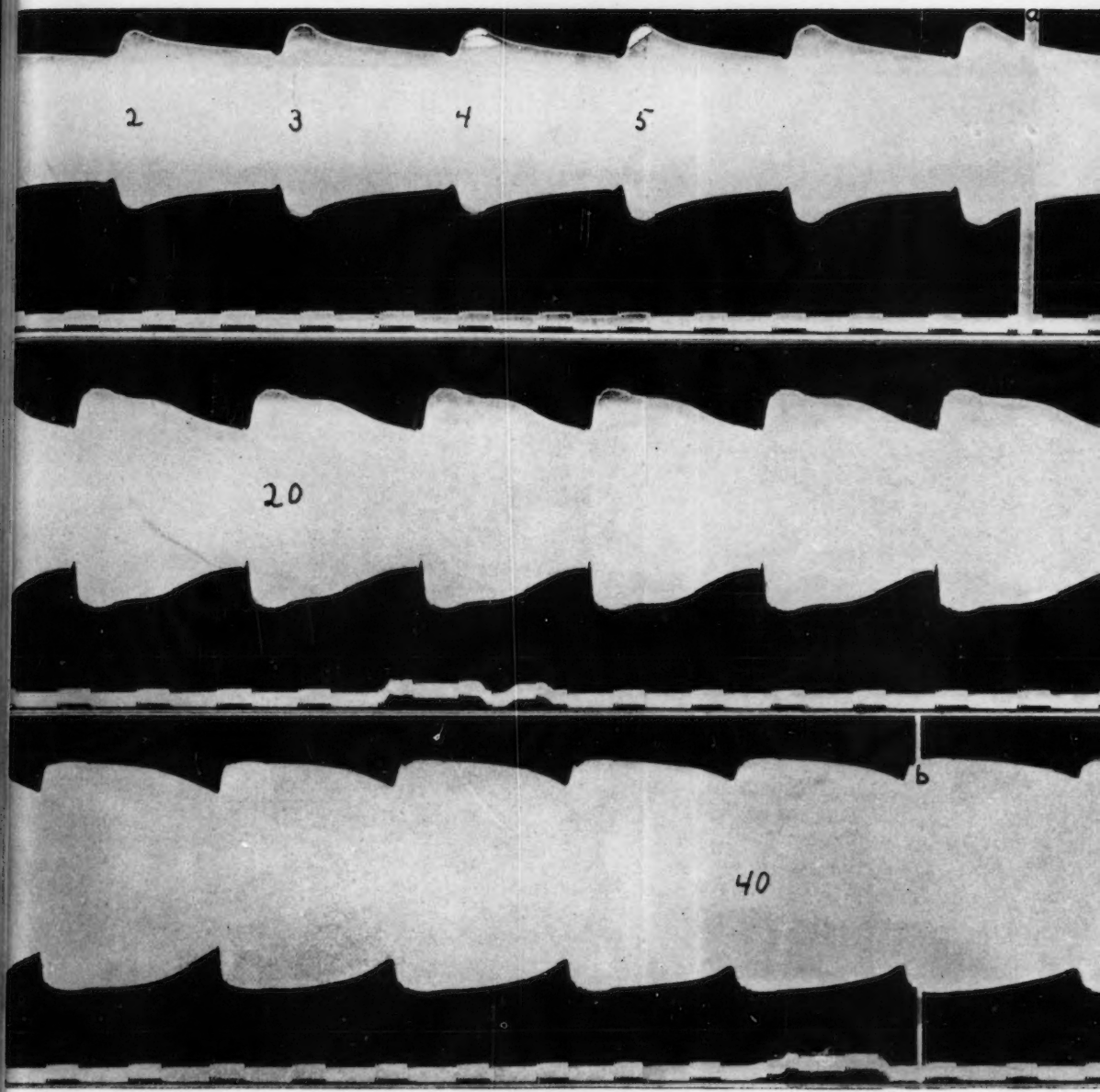
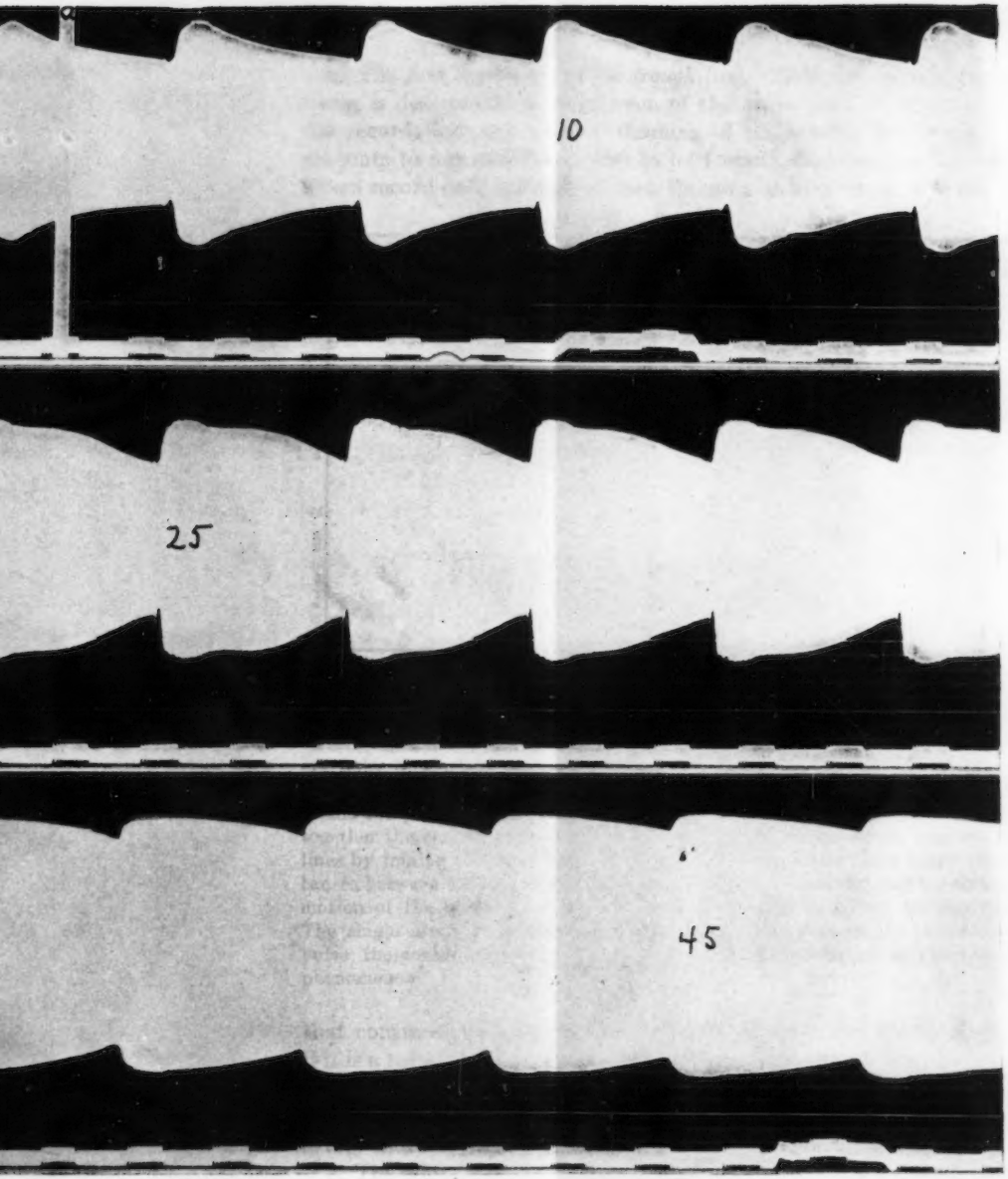


Fig. 8. A complete shadow record obtained from dog VI. Natural size. The shadow of the artery is inverted on the record. Owing to extraneous noises the signals, *a*
of the time



e record. Owing to extraneous noises the signals, *a* and *b*, intended to indicate sound phases, are valueless. Time in fifths of s
of the time record.



ne in fifths of seconds. Each pressure decrement of 10 mm. Hg. is indicated by the momentary elevation

a. The first depression of the trough line. This, the shadow records show, is due mainly to depression of the artery as a whole. Some of the records indicate a slight thinning of the artery, but at best this amounts to not more than 0.03 to 0.04 mm. Inasmuch as the mirror levers record only one-half of such thinning as may occur, it is obvious

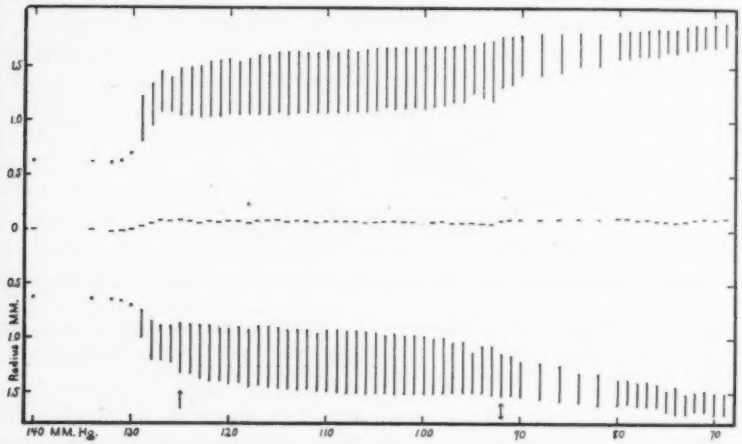


Fig. 9. Diagram of a shadow record obtained from dog VI. The lines show the radial amplitude of oscillation of the upper (top series) and lower (bottom series) edges of the artery as measured from the level of the end of diastole of one pulse (neglecting the pre-anacrotic phenomenon) to the crest of the succeeding pulse and set down according to scale, the zero being the center of the shadow of the completely compressed artery. The trough lines are formed by joining together the central ends of all of the lines of each of the series, and the crest lines by joining the outer ends. The points on the broken line bisect the distances between the two trough lines and are taken to indicate such up and down motion of the artery as a whole as may be occurring during decompression. The single arrow indicates the appearance of a new wave on the plateau of the pulse; the double arrow, the disappearance of the last vestige of the pre-anacrotic phenomenon.

that continued thinning of the flattened artery is not the main cause, if it is a cause at all, of the first depression of the trough line.

b. Second depression of the trough line. The shadow records (see figs. 9, 10 and 11) indicate that this is due entirely to depression of the artery as a whole.

c. The records also show the "*negative wave*" here under consideration, but on the upper surface of the artery only. In figure 8 this is first

seen in pulse -4. Inasmuch, however, as the lower surface of the artery at the same time exhibits a positive wave paralleling the negative wave, it follows that the negative wave as inscribed by the mirror levers is actually a pulsatile depression of the artery as a whole.

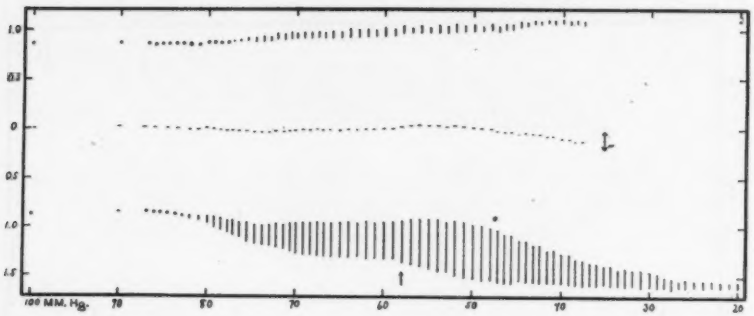


Fig. 10. Diagram of a shadow record obtained from dog VII. The upper surface of the artery seemed dry, and the compressed artery seemed to be bowed, rather than flat, on section. For further description see figure 9.

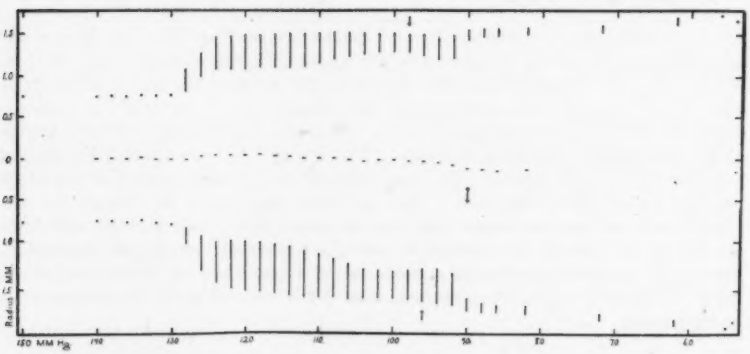


Fig. 11. Diagram of a shadow record obtained from dog VIII. For description see figure 9.

d. In pulse -3 and better in pulses -2 and -1, it can be seen that the artery thickens during diastole. This occurs during the period the mirror levers are recording the *first rise of the trough line*.

Discussion. On the basis of the foregoing observations, the changes in the level of the trough line occurring during the early stages of decompression can be satisfactorily explained if it is assumed that they are determined by a combination of *1*, depression of the artery as a whole, and *2*, the passage of blood along the artery during the diastolic phase of the pulse.

1. When the compressing pressure is thrown upon the artery the valves at the orifices close down on the artery and as they bulge outward they probably stretch the artery slightly between them. Any sag the artery in the chamber may have would thereby be diminished. It is also possible that the lower end of the artery sinks as a result of continued draining of blood from the artery beyond the occlusion. With decompression, the reverse process would take place; in this way a part of the slight depression that occurs during the early stages of decompression might be accounted for. The more rapid depression that occurs just before the pulse proper reaches a given lever presumably would be due to an acceleration of the sagging by the weight of the blood that now is entering the artery above. The "negative waves" that appear at about the same time, on this basis, probably would be an expression of a pulsatile sagging of the artery due to pulsatile weighting down of the artery with blood, to pulsatile elongation of the artery, and pulsatile shoving of the artery into the chamber. The fact that the lower end of the artery sinks further than the upper end, which indeed often does not sink at all, may be attributable to the slenderer proportions of the artery in the lower aperture of the chamber.

2. Presumably the diastolic increase in the caliber of the lower end of the flattened artery is due to the recession of a pulse that fails to get all the way through; some of the blood carried by the pulse remaining behind upon recession of the pulse and, becoming trapped in the compressed segment as its lumen diminishes to a narrow chink, finds, owing to the lower pressure peripherally than centrally, distad escape easier than centrad. The result is that blood, pushed by the compressing pressure, penetrates the compressed artery subsequent to the retirement of a pulse that penetrated the artery only partially. Such a layer of blood would be disclosed by the *levers* only when its thickness increases more rapidly than the artery simultaneously sags. At such a time the lever would be higher at the end of a given pulse cycle than it was at its beginning. Thus, presumably, is produced the remarkable picture of a pulse with an ascending diastolic phase.

Inspection. Simple inspection of the artery through the glass cover of the chamber furnishes interesting information relative to this discussion. The completely compressed artery, owing to its bloodlessness, is white in color. When blood enters during decompression, the artery can be seen to flush. If one compares the movements of lever *3* with the flushing of the artery at that point, it becomes quite clear that the artery flushes before the arrival of a pulse with a definite anacrotic limb, and that the flush then persists, though possibly becoming fainter, through the whole of that and of the succeeding diastoles.

In order to obtain more definite information with regard to this relation, in one of the experiments an observer watched lever *3* while another watched lever *1* and the color of the artery, and the compressing pressure was read when the former made out the first faint movements

TABLE 1

		MILLIMETERS OF Hg.			
Observer 1	Pulsation of lever 1	150	150	150	150
	Artery flushes	140	144	144	140
Observer 2	First movement of lever 3	140	144	150	140
	Brusque movement of lever 3	134	136	136	136

of the lever and the brusquer movements that occur when the anacrotic limb appears, and when the latter noted the pulsation of lever *1* and the first flushing of the artery at the level of lever *3*. Table 1 gives a few of the values thus obtained. These show that the reddening begins about 6 to 10 mm. Hg. after lever *1* begins to pulsate and practically at the time lever *3* begins to pulsate faintly; and that the two former events precede slightly (by about 4 mm. Hg.) the appearance of the brusquer pulses at the level of lever *3*.

The pictures obtained by means of both of the recording methods, therefore, are in agreement with each other and confirm the fact, made evident by inspection, that the artery flushes shortly before it is penetrated by the pulse and that then the flushing persists through the whole of the pulse cycle; the artery does not completely empty itself of blood during diastole once blood has succeeded in penetrating it.

DIASTOLIC EMPTYING OF THE ARTERY DURING THE LATER STAGES OF DECOMPRESSION

Mirror levers. In the midst of the events we have been describing the anacrotic limb of the pulse first becomes evident. Along with this there is, of course, a greatly increased systolic filling of the artery with blood, and therefore a much larger volume of blood to be expressed from the artery during diastole. The consequence is that during the diastolic phase the lever, as we have already mentioned in passing, now sinks to the next pulse, instead (in the case of the lower levers) of rising; that is to say, the wave now becomes typical of the form of a pulse wave. Nevertheless, for some pulses (*1* to *12*) the diastoles of the successive pulses continue to terminate on a higher and higher level—the trough line continues to rise. Here, presumably, a steadily increasing residue of blood is left in the artery at the end of each pulse; this residue presumably causes the lever to rise higher than the artery is depressed, if it is still depressed, under the weight of the residue.

It should be added that the elevation of the trough line that occurs at this time is exhibited by all the levers, even by those (lever *1*, most often) that do not record the second depression of the trough line. When there is no second depression, however, the elevation of the trough line may begin with the first pulse that penetrates into the compressed artery, or a pulse or two later. The trough line usually continues to rise relatively rapidly under all levers to about the same stage of decompression. There is some slight variability with regard to the time of onset of the rise relative to the penetration of blood and with regard to the time of termination of the rise relative to the stage of decompression, but not more than might be expected. For, assuming that our explanation of the rise of the trough line is correct, the balance struck between elevation of the lever by residual blood and depression of the lever by increase in weight (and other causes of translational movement) of the artery must differ at different points along the course of the artery. In no record has this phenomenon been missed; it cannot, therefore, be regarded as an artefact.

If measurements are made from the lowest level attained by the trough line during the early stages of decompression it is found that the more rapid rise carries the beam upwards through a distance that varies between 0.5 and 6.4 mm., the average of fifteen records being 3.4 mm. Disregarding for the present any possible added sagging of the artery that may be occurring, the thickness of the layer of blood

in the artery attained in this stage of decompression therefore ranges between ($0.5 \times 2 \times 0.04 =$) 0.04 and ($6 \times 2 \times 0.04 =$) 0.48, the average being ($3.4 \times 2 \times 0.04 =$) 0.27 mm.

A more or less sudden change in the direction of the trough line now takes place. As a rule it begins to sink slowly and more or less steadily, though it may run horizontally or even rise slightly. In general the tendency of the trough line to decline seems to be greatest under lever *3* and least under lever *1*. Indeed, the trough line written by lever *1* usually does not sink at all. One gains the impression that the smaller the artery the greater is the tendency for the levers to sink. And it seems to be the case almost without exception that the degree to which a given trough line now sinks varies with the depression that occurred just before the pulse succeeded in penetrating the artery. We believe, therefore, that the sinking of the trough line that is seen after the first rapid rise is a manifestation of the process that caused the first sinking; presumably this continues to act uninterruptedly, but its effect is masked temporarily early in the first-to-fourth phase period by the relatively rapid accumulation of a diastolic residuum in the artery. From what has been said above, it would seem that the amount of the depression depends upon the caliber of the artery with respect to the orifice of the chamber; the larger arteries would be better supported at the orifices of the compression chamber, and so also would be the upper end of a given artery, for there it is larger than at the lower end.

This more horizontal phase of the trough line is terminated by a very obvious turn upward which usually begins in the latter part of the first-to-fourth sound-phase period. It seems fair to assume that those records in which the trough line at no time sinks indicate more accurately the changes in the amount of diastolic residuum occurring in this stage of decompression during which the trough line runs approximately horizontally. In view of the fact, therefore, that the trough line in this stage either is horizontal or sinks, it may be concluded that after the termination of the first rapid increase in diastolic residuum that occurs at the time the pulses first penetrate there is for some time no, or at best only a very slight further increase in the amount of the diastolic residuum. Selecting that part of the trough line that lies between the close of the first rapid rise (usually in the vicinity of pulse *3* or *4*) and the beginning of the second rise, in records which at no time give evidence of a gravity depression, it is found that the maximum total recorded rise of the trough line in this phase amounts to about 1 mm., or a further increase in the thickness of the artery amounting to only

$(1 \times 2 \times 0.04 =)$ 0.08 mm. In view of what has been said this must be regarded as a minimum figure.

As has been said, after a considerable part, usually about two-thirds, of the first-to-fourth sound-phase period has run its course, the trough line begins to rise, or, if it has been rising, the rate of rise increases appreciably. This rise does not begin at the same time under all of the levers; and there seems to be no regularity with respect to the part of the artery in which it first appears. In the vast majority of instances the rise at first is slow but soon a striking acceleration develops. Eventually this accelerating rise changes more or less abruptly into a retarding rise. During the accelerating rise the steps from pulse to pulse may be very irregular, the trough line not infrequently zigzagging to the top of this phase. These irregularities often make it difficult to interpret the records. The subsequent slower rise, which at first also may be

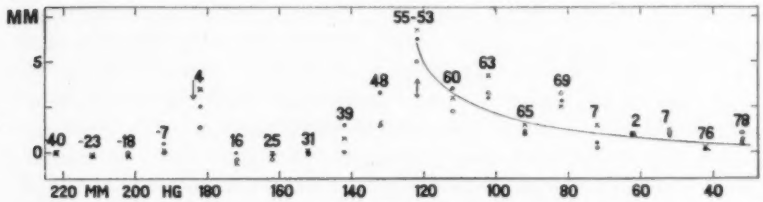


Fig. 12. Diagram constructed from figures 4 and 7 to bring out the rate of change of the diastolic level.

somewhat irregular, apparently follows a hyberbolic curve which starts from the accelerating rise usually at a very noticeable angle and tends to become parallel to the abscissa. This inflection of the curve of rise usually affects all of the levers in about the same pulse cycle, namely, the cycle, or at most one or two cycles removed from the one, in which the beginning of the fourth sound-phase is signalled and also the cycle in which the pre-anacrotic negative wave, to be considered below, suddenly disappears, or begins to diminish rapidly in amplitude in those instances where it does not suddenly disappear.

In figure 12 are plotted the increments or decrements in the level of the trough line in mm. for each fall of 10 mm. Hg. of the compressing pressure as recorded in the case of dog V. It will be recalled that in the experiment that furnishes the data for this curve the compressing pressure was recorded by signalling each fall of 10 mm. Hg. We have selected for the construction of the curve those pulses that coincide

most closely with each of these signals. By thus including in each step of the curve a considerable number of pulses the irregularities from pulse to pulse are largely eliminated as long at least, as the compressing pressure is falling slowly. But when the compressing pressure is falling so rapidly that only 2 or 3 pulses record between successive signals, as is the case toward the close of this record (below 112 mm. Hg.) the failure of the signal to always appear in the same phase of the pulse cycle that happens to lie over it, while not altering the general form of the curve causes considerable irregularity to appear in the position of the points around the curve. It should be added that the rate of the fall of the compressing pressure was increased only after the fourth sound-phase had been signalled and in order not to protract unnecessarily the record, which, as it was, attained a length of 4.5 M.

This curve brings out clearly the accelerating elevation of the trough line as the fourth sound-phase is approached and the retarding elevation, very distinctly hyperbolic in form, thereafter. The point of inflection agrees exactly with the pulse in which the fourth sound-phase begins and the pre-anacrotic phenomenon disappears. The agreement in other cases is not always so close. Indeed, the exactness of the agreement in this case is due in some, though small, measure to the large size of the steps of the curve (the decrements are 10 mm. Hg.). If smaller decrements, for example, pulses, had been chosen the agreement would not have turned out quite so close.

To summarize, the trough line during decompression, especially as written by levers *2* and *3*, at first sinks slowly, then more rapidly for a few beats just before the pulse reaches the respective lever. With the arrival of the pulse at the position of a given lever the trough line written by it rises rather rapidly for a few beats, 3 to 11 or more, this rise taking place progressively down the artery. Lever *1* then usually writes a horizontal or slightly rising line; levers *2* and *3* a slowly sinking line. After decompression has proceeded, usually considerably more than half of the way through the first-to-fourth sound-phase period, all of the trough lines begin to rise, usually slowly at first, and then in leaps and bounds. At the time the beginning of the fourth sound-phase is signalled, and at about the time the pre-anacrotic negative wave disappears, this accelerating rise changes more or less rapidly into a retarding, though still relatively rapid rise.

It is difficult to interpret these results in terms of thickness of the layer of blood remaining in the collapsing artery at the end of diastole. For, as has been made clear, the depression of the levers probably is due to depression of the artery as a whole. There is no safe way of

making an allowance for this factor. Then again, as will be seen later on, distention of the artery peripherally may cause the artery, especially at its lower end, to rise in the compression chamber; and it is quite likely, therefore, that when blood begins to freely penetrate the chamber and to distend the artery beyond, the sinking of the lower end of the artery may give way to elevation. But as it is important to have some, even though rough, idea with regard to the changing thickness of the layer of blood in the artery, we have essayed to measure it in three of the records (those plotted as figs. 5, 6 and 7). In making these measurements we have gratuitously assumed *1*, that the rate of depression of the artery is uniform, and corresponds with the early rate of depression, as long as the lever sinks at all; *2*, that as soon as the lever begins to rise the artery no longer sinks but maintains its position through the remainder of decompression; and *3*, that where the lever does not at any time sink, the artery at no time sinks. From base lines derived through these assumptions, the thickness of the layer of blood has been derived. The data are collected in table 2. They will be discussed after consideration of the changes in diastolic bore of the artery as determined by the shadow method.

Shadow records. For reasons already discussed, the shadow records of decompression are practically uncomplicated by translational movements of the artery; therefore the changes that occur in the thickness of the artery at the end of diastole can be read off of these records directly. It should be recalled, however, that failure of the artery when compressed to become quite flat and in the horizontal plane will, until the artery becomes full enough to permit the illuminating rays to become tangent to the artery at the lines formed where the vertical plane passes through the axis of the artery, have the effect of masking the magnitude of the earlier changes in the thickness of the residual layer of blood. It should also be borne in mind in this connection that whereas in the case of the femoral artery the peripheral emptying of the segment of artery in the chamber is relatively unobstructed until decompression reaches the point where the flow becomes fairly free, in the case of the carotid artery used in the shadow chamber the emptying will always occur against a relatively high peripheral pressure. Some of the differences of the manner in which the trough line rises as between mirror and shadow records presumably are attributable to these differences in conditions.

The figures (9, 10 and 11) and table 3 show at a glance that the changes in residual blood are qualitatively the same as those made out by the labored interpretation of the records obtained by means of the

TABLE 2

INCREASED THICKNESS OF ARTERY FROM LOWEST LEVEL TO	LEVER	DOG III				DOG IV				DOG V			
		Duration of phase in pulses	Rise above collapse (a)	Indicated thickness (a × 2 × 0.04)	Per cent of total	Duration of phase in pulses	Rise above collapse (a)	Indicated thickness (a × 2 × 0.04)	Per cent of total	Duration of phases in mm. Hg.	Rise above collapse (a)	Indicated thickness (a × 2 × 0.04)	Per cent of total
			mm.	*mm.*			*mm.*	*mm.*			*mm.*	*mm.*	
End of first rise.....	1	−7 to 8	3.2	0.256	29.4	−6 to 3	2.0‡(?)	0.160	9.4(?)	197 to 182	6.4	0.512	11.0
	2					−2 to 3	5.4	0.432	23.5	188 to 182	8.0	0.640	13.8
	3	1 to 3	2.0	0.160	10.5	−1 to 2	3.2	0.256	24.6	185 to 182	4.4	0.352	9.2
Beginning of second rise*.....	1	8 to 35			29.0+(?)	−3 to 31−	3.2(?)	0.256(?)	15.0(?)	182 to 146		0.512	11.0(?)
	2					3 to 31(?)		0.432	23.5(?)	182 to 146		0.640	13.8(?)
	3	3 to 31			10.5(?)	2 to 35		0.256	24.6(?)	182 to 146		0.352	9.2(?)
End of accelerating rise**.....	1	35 to 50	8.0	0.640	73.4	31 to 40	16.7	1.336	78.1	146 to 120	29.0	2.320	50.0
	2					31(?) to 40(?)	17.2	1.376	74.8	146 to 120	38.0	3.040	65.5
	3	31 to 52	12.2	0.976	64.3	35 to 40	11.0	0.880	84.7	136 to 120	28.0	2.240	58.5
End of record[a].....	1	50 to 61	10.9	0.872	100.0	40 to 46	21.4	1.712	100.0	120 to 78	58.0	4.640	100.0
	2					40 to 46	23.0	1.840	100.0	120 to 78	58.0	4.640	100.0
	3	52 to 61	19.0	1.520	100.0	40 to 46	13.0	1.040	100.0	120 to 78	47.8	3.824	100.0

* It is assumed that no increase in thickness occurs in the case of those records which in this stage run horizontally or sink, and that when the lever sinks the decline proceeds at a uniform rate from the beginning.

** It is assumed that the artery stops sinking in this and the subsequent stages.

‡ Taken from another record starting at a higher compressing pressure.

[a] Decompression is never complete at this time, and is not the same in different observations.

TABLE 3

Shadow records; thickness of artery reduced to actual size

	DOG VI					DOG VII					DOG VIII				
	Pulse number	Diastolic thickness	Thickness of layer of blood	Per cent of total diastolic increase	Compressing pressure	Pulse number	Diastolic thickness	Thickness of layer of blood	Per cent of total diastolic increase	Compressing pressure	Pulse number	Diastolic thickness	Thickness of layer of blood	Per cent of total diastolic increase	Compressing pressure
		mm.	*mm.*		*mm. Hg.*		*mm.*	*mm.*		*mm. Hg.*		*mm.*	*mm.*		*mm. Hg.*
At beginning of record		1.78			140		1.74			100		1.50			140
At beginning of first rise	−3	1.78			134	−4	1.71			86	−3	1.51			134
At end of first rise	3	1.97	0.19	14.3	127	11	1.87	0.16	17.2	75	3	2.09	0.58	27.2	124
Irregular rise begins	23	2.02	0.24	18.0	107	34	1.89	0.18	19.2	58	6	2.09**	0.58	27.2	118
											19	2.60**	1.09	51.2	92
Pre-anacrotic wave gone	38	2.48	0.70	52.3	92	56*	2.46	0.75	79.8	35	20	3.13	1.62	76.1	90
End of accelerating rise	38–40	2.64	0.86	64.8	90	52–53	2.50	0.79	84.0	34					
End of record‡	55	3.11	1.33	100.0	70	73	2.65	0.94	100.0	20	38	3.64	2.13	100.0	54

* But with pulse 52 there is a very sharp diminution in the pre-anacrotic phenomenon.

** In this case a regular rate of increase showing respiratory waves begins with pulse 6; with one pulse (19–20) a tremendous increase takes place, and then a more gradual rate of increase is resumed.

‡ Decompression is never complete at this time, and is not the same in different observations.

mirror levers. As in the case of the mirror records, there is in this case also, considerable variation from animal to animal. In dog VI (figs. 8 and 9) the diastolic thickness of the artery increases rapidly from pulse *−3* to pulse *4*. With these pulses, subtending 7 mm. Hg. of decompression, 14.3 per cent of the entire diastolic increase occurs. The increase occurring during the next 20 pulses, subtending 20 mm. Hg., amounts to only 3.7 per cent of the total. This practically level stage is then succeeded by a stage lasting 17 pulses and 17 mm. Hg. of decompression in which the thickness of the artery increases rapidly and irregularly; 46.8 per cent of the total increase occurs in this stage. During the remainder of the decompression (through 20 mm. Hg., lasting 15 pulses) the rate of increase becomes regular again and, though relatively rapid, tends constantly to become slower.

The upper surface of the artery in the case of dog VII (fig. 10) had dried somewhat and, therefore, did not move as freely as the lower. In this case the first increase in thickness lasts through many more pulses, *−4* to *11* and through 11 mm. Hg., the increase in this stage amounting to 17.2 per cent of the total. During the next 23 pulses the increase amounts to only 2.0 per cent, though the compressing pressure falls 17 mm. Hg. This is succeeded by the stage of rapid, irregular increase which, in the course of 24 pulses and a decrement of 24 mm. Hg., brings the thickness up to 84.0 per cent of the total attained 15 pulses and 15 mm. Hg. later, where the record ends.

In the case of dog VIII (fig. 11) the first rapid increase in thickness occurs during 6 pulses, *−3* to *3*, and 10 mm. Hg. decrement and makes up 27.2 per cent of the total attained. The stage of slower increase in this case is represented by only 3 pulses. It is perhaps more correct to say that there is no such stage in this case; that, instead, the thickness increases in regular waves, probably respiratory in origin, through 16 pulses and 32 mm. Hg. to acquire in this time 51.2 per cent of the total. Then in a single pulse cycle, *20*, an increase of 24.9 per cent occurs and this is succeeded again by a slower and steady increase from the 76.1 per cent to the 100 per cent line in the course of 18 pulses and a further compression fall of 28 mm. Hg.

The shadow records, therefore, show that during decompression blood begins to penetrate before the pulse, and that after blood has once entered the artery, the latter does not again completely collapse during diastole. This diastolic residuum at first accumulates rapidly so that usually before the decompression has proceeded 10 mm. Hg. after the blood has succeeded in penetrating to a given point, it has acquired an

actual depth of 0.2 mm., in round numbers. Occasionally greater depths, up to 0.6 mm., have been noted. The cause of such differences is not clear; possibly the presence or absence of partial obstructions to the emptying of the compressed segment offered by fortuitous contacts of the artery with the orifices of the chamber accounts for them. Then, with rare exceptions, only a very slow further increase in residual blood occurs until the decompression has proceeded well beyond the mid first-to-fourth sound-phase period, when a rapidly accelerating diastolic accumulation occurs. The larger part of the accumulation of this stage may be accomplished in one or two pulse cycles. At other times it runs through a larger number of pulses. At about the time the fourth sound-phase begins this gives way to a retarding, though still rapid, increase in diastolic volume.

If now we analyze more closely the results obtained by the aid of the mirror levers, as shown in table 2, upon the basis of the assumption made on page 111, it is found that not alone is there a close qualitative agreement as to the way in which the diastolic residuum accumulates, the agreement, indeed, is almost quantitative. Thus the first rapid accumulation of the diastolic residuum averages under all levers 16 per cent of the total diastolic caliber attained during decompression; while by the shadow method the corresponding figure is 19 per cent. The change in diastolic caliber that occurs during the relatively level stage cannot be estimated in this case from the mirror lever records. But that occurring during the subsequent accelerating rise can; it averages 69 per cent of the total. The corresponding value derived from the records made by the shadow method is 61 per cent. These values can therefore be accepted as indicating roughly the rate at which the diastolic residuum accumulates during decompression.

Previous observations on diastolic residuum. In Part I (2) of this series it was argued that in a pulsating circulation schema and, under ordinary circumstances, in animals also, where the geometric mean of the pulse waves lies to the diastolic side of the arithmetic mean of the systolic and diastolic pressures, and allowing for the resistance of the arterial wall to deformation, the artery probably would completely collapse during diastole as long as it was under a compressing pressure that exceeded the diastolic pressure. To the eye this was exactly what happened in the experiments with the model employed in that investigation, in which the diastolic period was long (several seconds) and the rise of pressure producing the pulse very brief. With a circulation model employed in the present experiments the same fact has

again been demonstrated, this time, however, with the aid of the delicate mirror levers. When the diastolic period is long (several seconds) the artery practically collapses in diastole as long as the compressing pressure exceeds the diastolic pressure, allowance, of course, being made, as always has been the case, for the resistance to compression offered by the arterial wall. At the same time the present experiments have shown that for the attainment of complete or practically complete collapse during diastole, the diastolic phase must last a very much longer time than we had believed to be necessary.

An experiment simulating the one referred to in the preceding paragraph designed to determine the time required for the complete emptying of the artificial artery has been devised with the artery in situ in the compression chamber. In an animal the artery first was clamped beyond the stethoscope, then central to the compression chamber, and the pressure in the compression chamber then raised until it exceeded the pressure necessary to completely collapse the artery. Under these circumstances, of course, the artery remained full. Then with a stopwatch the time was determined that elapsed between opening the lower clamp and the termination of the obvious movement of the beam of light reflected from the mirror of lever *3*. This was found to be about two seconds. This then, roughly, is the time necessary for the complete emptying of the compressed segment under a compressing pressure that exceeds the systolic pressure, when the peripheral pressure is as low as it will fall, but when emptying in a central direction is prevented. The period is much longer than that of a normal pulse cycle.

Also in Part I of this series (2), some observations were made on the volume changes of the artery in the chamber during decompression. An attempt was made to explain these changes in diastolic volume upon the assumption that the parts of the artery well within the compression chamber completely empty themselves during diastole until the fourth sound-phase begins. Upon this assumption it seemed possible to attribute all of the changes in diastolic volume occurring before the accelerating increase that begins late during compression, to the downward and then the upward extension of the upper and lower conical closures, respectively, of the artery. The uncertainty of the assumption utilized as the basis of this analysis was one of the reasons for the development of the present methods of studying the movements of the artery within the compression chamber. The results now obtained conclusively invalidate the earlier assumption. When, however, the results obtained in the present and the previous studies are analyzed

side by side it becomes clear that they actually supplement each other beautifully even to the point of demonstrating that during decompression the artery retains blood before the pulse proper begins to penetrate.

Thus, taking the figures from table 4 published in connection with the earlier observations and comparing them with figure 18 in the same place (2), it is found that at the time the volume pulse begins to increase in size (pulse *4*) the diastolic volume increases rather rapidly, and until, with pulse *8*, the volume of the arterial contents in diastole amounts to 0.033 cc. If it is assumed that the width of the artery when pressed flat is 3 mm. the thickness of the diastolic layer of blood at this time, if uniform, would be 0.22 mm., a figure that compares favorably with those obtained by calculation from the mirror and shadow records of the present paper.

As in the case of the shadow and mirror records, this stage of rapid increase is followed by a stage of slow increase extending from pulse *9* to about pulse *31*, inclusive. The increase in this stage amounts to only 0.0247 cc., and the diastolic layer of blood at its close, determined as above, by calculation, has a thickness of 0.38 mm.

Now the diastolic volume begins to increase at an accelerating rate; the largest step is in association with pulse *38*, after which the contents of the artery amount to 0.1897 cc. At this time the artery is no longer flat but nearly or quite cylindrical. The thickness of the column of blood, determined by calculation, is 2.2 mm.

It is thus seen that not alone does the diastolic volume of the artery, directly measured, increase in the same fashion as the diastolic residuum as determined by means of the mirror and shadow records, but in addition the values as determined by calculation from the data derived through all of the methods are in as close agreement as could be expected when allowance is made for the large inherent experimental errors. It becomes clear, therefore, that in our previous papers we greatly over-estimated the importance of the extension of the upper and lower conical closures of the compressed artery.

CONFIGURATION OF THE DIASTOLIC PHASE OF THE PULSE IN RELATION TO DIASTOLIC RESIDUUM

The theory of the diastolic emptying of the compressed segment of the artery has been considered; and it now remains to be seen in how far theory is in accord with the facts and in how far it accounts for the manner in which the diastolic residuum increases during decompression.

Turning to the records, it is seen that at the time the pulse begins to penetrate the length of the artery the diastolic phase has the configuration of pulse *172 a* in figure 2; the diastolic phase tends to, but does not actually, become parallel to the base line. Obviously emptying is proceeding throughout the pulse cycle but, toward the close, at a very slow rate, and there is not time in the pulse cycle for it to attain completion. In this fact, evidently, is to be found the explanation of the first rapid rise in the level of the trough line.

Furthermore, it is seen that the diastoles preserve this configuration for a considerable distance through decompression, especially those recorded at the lower end of the compressed segment. Despite the increased systolic filling that occurs during decompression and the consequent increase in the amount of blood to be emptied during diastole, notwithstanding, even, the constantly decreasing compressing pressure, the diastolic residue now accumulates only very slowly. The diagram (fig. 2) indicates why this is to be expected. It is there seen that owing to the form of the curve of emptying the termination rises very little during the first 40 mm. of the fall of the compressing pressure below the systolic arterial pressure, a value which amounts to about two-thirds of the pulse pressure.

As decompression proceeds, this more horizontal phase of the diastole as recorded can be seen to gradually shorten, and the second rapid rise of the trough line begins when the after-coming pulse arrives during the earlier and steeper parts of the curve of collapse. Some resemblance of the pulses recorded at this time to curve *132* (fig. 2) can usually be made out. A sudden change from the rapid descent of the diastoles of this period to a more gradual descent often is seen (see fig. 4), and it then corresponds accurately with the disappearance of the pre-anacrotic phenomenon and with the termination of the accelerating rise of the trough line. But in many instances this sudden change is not discernible.

The only explanation of this variation that has occurred to us is based upon the effect occlusion of the artery beyond the chamber has upon configuration of the terminal parts of the diastolic phase. When, in dog IV, the artery is thus occluded before it is compressed, it is found that during decompression lever *3* at no time shows the late steep descent during diastole; and this feature becomes much less prominent at the position of levers *1* and *2* also. In the case of dog V, occlusion of the artery peripherally when the decompressing pressure is held constant at a time when the steep diastolic descent is recording

causes the descent to disappear under lever *3*, and if anything to become more pronounced under levers *1* and *2* (see fig. 13). It would seem, therefore, as might have been anticipated, that the facility with which the artery empties itself peripherally in the stage in which blood is passing freely, is an important factor in the rate with which the artery collapses.

The diagram (fig. 2) shows why we should expect to obtain a rapidly accelerating accumulation of a diastolic residuum at this time (pulses *142* and *137*). It also makes clear how, in this stage of decompression, very slight changes in the arterial pressure or in the duration of the pulse cycles will result in the wide fluctuations in the amount of the diastolic residuum that are observed in this stage of the experiments.

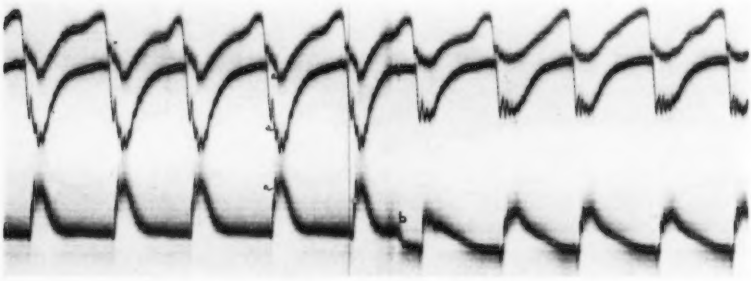


Fig. 13. Part of a mirror lever record from dog V, showing the effect of de-occluding (at *b*) the artery peripheral to the chamber while the compressing pressure is held constant at a relatively low level, but within the critical stage. Reduced.

Finally, the diagram shows that the accelerating accretion to the diastolic residuum should give way rather abruptly, as it does, to a retarding increase and that this change occurs at the moment the artery just fails during diastole to collapse below its just round size. This must, of course, occur at about the time the compressing pressure falls below the intra-arterial diastolic pressure.

THE SYSTOLIC PHASE

Progression of the pulse into the artery. The study of the systolic phase can best be begun with a consideration of the phenomenon that has given rise to the least difficulty in interpretation, namely, the

progression of the pulse into and through the compressed artery. Under compressing pressures which unquestionably are high enough to completely stop the pulse, the levers may show slight movements with each of the pulses. These movements, which are always most obvious in the record written by lever *1*, and least obvious, indeed, usually entirely absent, in the records written by lever *3*, have been seen when the compressing pressure has exceeded the systolic pressure by as much as 40 mm. Hg. That they are not due to the penetration of blood down to the lever exhibiting the motion is proved by the fact that the artery as viewed at the time through the glass window is quite flat and white, and exhibits no pulsatile flushing such as is evident when even the thinnest layer of blood begins to penetrate.

Probably a number of processes combine to produce this *preliminary pulse*. One factor may be the movement imparted to the artery through chance contacts between it and the central orifice of the chamber. To this possibility is to be added the effect of the impact of the pulse against the central conical closure of the artery. The fact that the preliminary pulse usually is a miniature of the fully developed pulse is in keeping with these possibilities. But whatever the cause of the preliminary pulsation may be, it is certain that it is very remotely, if at all, related to any of the significant processes enacted in the compression chamber.

During decompression the advent of the pulse proper to the position occupied by a given lever usually is clearly indicated by the appearance of an anacrotic limb, or of a new peak on the record. At the time this first appears under lever *1*, there is no evidence of it under levers *2* or *3*. With further decompression, the pulse proper soon appears under lever *2* and then under lever *3*. A few of the values illustrating the rate of the increase in the depth of penetration of the pulse with decompression are collected in table 4. The table gives the number of pulses elapsing, and the estimated pressure decrement, between the appearance of the clear anacrotic limb at any two of the levers. Owing to the absence of a simultaneous record of the rate of fall of the compressing pressure in the case of dogs III and IV, and owing to the possibility of some lack of uniformity in the rate of decompression, the assigned pressure differences in these instances can be regarded only as very rough approximations; the figures given in connection with dog V, however, are correct to within 1 mm. Hg. Furthermore, rather wide variations are bound to occur from record to record as a result of the changing relation of respiratory and other waves of pressure to the

compressing pressure. Taking the figures as they stand, the table shows that the pulse proper appears later under lever *3* than under lever *1* by from 6 to 15 pulses and by a pressure decrement averaging 14 mm. Hg. As the distance between these levers is 51.5 mm., it follows that after the pulse during decompression begins to enter the compressed segment the further penetration of the pulse into the artery proceeds roughly at the rate of 1 cm. per 2.7 mm. Hg. pressure decrement. The rate of penetration, though, is not constant, but seems to increase with the distance. Thus in every estimation save one, and in that case the number of pulses is the same, more pulses elapse during the penetration from lever *1* to lever *2*, a distance of 21.6 mm., than

TABLE 4

DOG NUMBER	RECORD NUMBER	DECREMENT REQUIRED TO PERMIT THE PULSE TO PROGRESS					
		From levers 1 to 3		From levers 1 to 2		From levers 2 to 3	
		Number of pulses	mm. Hg.	Number of pulses	mm. Hg.	Number of pulses	mm. Hg.
III	2	11	16–	6	9–	5	7–
	3	15	22–	8	12–	7	10–
IV	1 A	6–7	9–10	3–4	4–6	3	4
	2	6	9	3?	4.5	3	4.5
V	2	12	12	9	8	3	4
	3	11	12				

from lever *2* to lever *3*, a distance of 29.9 mm. No effort has been made to determine whether the rate is affected by changes in the conditions under which indirect blood pressure estimations are made.

A pulse model constructed to control this, as well as certain other, observations gave qualitatively similar results. When the pulse, produced by permitting a stopcock in the model to fly open, begins to penetrate the compressed excised artery, it does not at first course the entire length of the artery. In order to have it penetrate the length of the artery subtended by the two mirror levers, in these experiments a distance of 61 mm., it was necessary to lower the compressing pressure about 7.5 mm. Hg. In other words, the penetration in this case is at the rate of 1 cm. per 1.23 mm. Hg. of decompression; it, therefore, is faster than in the animal. So many of the conditions were different, such as pressures, coefficient of elasticity, form of pulse, viscosity

(water being used in place of blood), caliber of artery, etc., that the cause of the difference in the rate of penetration cannot be determined. As from the animal experiments, so too from the model, the impression is gained that the ability of the pulse to plow its way through the compressed artery increases with the distance it penetrates.

This observation is of some significance in relation to the problem of the effect of the width of arm band upon the values obtained in pressure estimations in man and in relation to the criteria to be employed in reading the systolic pressure. It has long been known from observations in man that increasing the width of the armlet lowers blood pressure readings. von Recklinghausen (7) pointed out that this effect might be due either to a tendency on the part of the wider cuffs to minimize tissue resistance or to a loss of pressure in the artery under the cuff as a result of the resistance the compression offers to the moving pulse (*"Wegebahnung"*). It was argued that if this property of *penetrativeness* were a factor the rate at which the pressure readings is lowered should increase with the width of the cuff. As a matter of fact, however, just the reverse has been found to be the case (1). It was therefore concluded that *penetrativeness* is a negligible factor in the blood pressure observations. In the present experiments, however, tissue resistance is not a factor; for we have determined that only a few millimeters Hg., usually not more than 2 or 3, are required to deform the part of the artery included between the two end levers, both of which lie something like 15 mm. within the compression chamber. And yet it is obvious that if the compression chamber had extended only to lever *1*, the systolic pressure readings made by the method of palpation, for example, would have been higher by over 10 mm. Hg., than were those that were obtained with the whole length of the chamber.

Penetrativeness, therefore, is a factor that must be reckoned with in blood pressure observations in man. It is difficult, however, to know how to allow for it. We have at present no information with regard to the length of artery that is occluded by the usual armlets under a compression that just counterbalances the arterial systolic pressure. If it is assumed that it amounts to only 4 cm., a minimal figure, and that the penetrability of the brachial artery of man to the pulse is the same as that of the femoral artery of the dog, it would follow that in man the systolic blood pressure readings, disregarding the error due to tissue resistance, are at least 8 mm. Hg. too low. But there seems to be no way of escaping the dilemma of the errors due to tissue resistance and to penetrativeness, so as to allow for these disturbing factors in routine observations of the arterial pressure.

It is obvious that the criteria of the systolic pressure that depend upon the penetration of the pulse into the peripheral artery, such as palpation and auscultation beyond the arm band, and probably the change in the form of the pulse (12) also, which we have found to coincide, to the pulse wave, with the return of the Korotkoff sounds, must give lower readings than any there may be that indicate when the pulse is beginning to overcome the resistance offered by the compressing pressure. Whether or not we are in possession of any criterion of the latter event is not quite certain. We are inclined to believe, however, that the criterion suggested in 1904 (1) namely, "a more or less abrupt increase in amplitude" is such a one. This criterion we subsequently abandoned for "the change in form" because a *sudden* increase in amplitude is rarely discernible. The reason for the absence of an abrupt increase now becomes clear. It is due to the fact that the pulse penetrates the length of the compressed segment gradually. Now, although under the circumstances no *sudden* increase in amplitude could be expected, we nevertheless should expect, when the pulse begins to penetrate the compressed segment, a rapidly accelerating increase in the amplitude of oscillation. Upon going back to our original records (see figs. 6 and 8 in that place) we find that this is exactly the criterion of the systolic pressure that was employed.

Taken by itself the question as to which criterion should be employed in reading the systolic pressure is of little significance just so long as the criterion employed is always the same and only the systolic pressure is determined. But when the systolic and diastolic pressures are taken together for the purpose of determining the pulse pressure it is very obvious that the two readings should be comparable. Inasmuch as observations recorded later in this paper indicate that the usual criteria of the diastolic pressure, neglecting the effect of tissue resistance, are not influenced by the length of the segment of artery compressed, it would seem that the preferable systolic criteria should be such as are not affected by the length of the segment. Final judgment on this question, however, must await further investigation.

Just why the rate of propagation of the pulse should increase, as it seems to, with the distance the pulse penetrates, is not entirely clear. Possibly the phenomenon has some relation to the increase in the steepness of the wave front that develops in association with the pre-anacrotic phenomenon as the wave progresses along the artery. If it is a fact, it would account for the observation (1) that the decrease in systolic pressure readings that occurs as the width of the arm band is increased diminishes as the width increases.

SYSTOLIC FILLING; THE CREST LINE

Mirror levers. In the measurement of the maximum systolic bore the same difficulty is encountered as was met with in the case of the measurement of the diastolic bore, namely, that of estimating and allowing for translational movements of the artery. The artery under lever *1*, as has been seen, usually is depressed but little through the action of gravity; therefore, the maximum radius of the artery attained during systole is fairly accurately indicated by the record made by that lever. But in order to derive from the records the largest radii of the artery under levers *2* and *3* it is necessary to take depression of the artery as a whole into consideration. This, as has been indicated, is apt to introduce a very large error. Yet measurement of the records brings out some features that recur so constantly as to give to them considerable credence.

In every case and under all of the levers the curve, the *crest line*, formed by plotting height attained by systole against compressing pressures (or pulses) has essentially the same configuration (see figs. 5, 6, 7). For some pulses after the pulse, during decompression, has reached a given lever the crest points on the curve mount rapidly and irregularly. These points, usually, can be grouped about a straight line. In any event, the crest line soon turns more or less sharply toward the horizontal; here, allowing for depression of the artery in the case of levers *2* and *3*, the curve is distinctly hyperbolic in form and the points fluctuate much less widely. Later, well along in the first-to-fourth sound-phase period, the crest line mounts again, rapidly at first, and then more and more slowly. The curve here, again, is usually quite regular and usually hyberbolic in form. This second rise begins earlier under lever *3* than under levers *2* and *1*.

Without going into too great detail, it may be said that the first rapid rise of the crest line terminates roughly, allowance being made for depression of the artery, within the range of the last step of the accelerating rise of the trough line as recorded by the same lever toward the close of the first-to-fourth sound-phase period. The mechanism of these two inflections presumably is the same; but of this more in a moment.

The second rapid rise in the crest line is due to the growth of a wave that appears at the time on the plateau of the pulse wave. All of the facts seem to indicate that the wave that produces this rise travels backward from the part of the artery beyond the compression chamber.

Thus it develops at a time when blood presumably is passing through the compressed segment at a relatively rapid rate during the whole of the cardiac cycle, and is maintaining a comparatively high pressure and tense walls peripherally, conditions that are favorable to a partial reflection of the primary pulse wave. Furthermore, the wave appears first under the more distal levers. Thus in figures 4 and 7 it is seen that it first appears above the level of the plateau of pulse *38* as written by lever *3;* under lever *2* it is first seen as a peak in pulse *41;* and under lever *1* it is first seen, again as a peak, on pulse *44*. When the wave is in evidence under all of the levers measurement shows, in other records more clearly than in the one here used for purposes of illustration, that in any given pulse cycle the wave begins earlier under lever *3* than under lever *1*. With decompression the wave moves forward on the pulse from cycle to cycle until its front finally fuses with the anacrotic limb of the primary pulse. The fusion is complete at about the time the artery during diastole fails to collapse below its just round size.

But the best clue to the nature of this wave is furnished by records that were made while occluding and deoccluding the artery just peripheral to the compression chamber while maintaining a constant compressing pressure in the chamber. It so happens that this observation was made on the dog from which figure 4 also was obtained. The compressing pressure was set at a relatively low level in the first-to-fourth sound-phase period; the Korotkoff sounds were loud and clear. With deocclusion (see fig. 13), which in this case gives clearer results than occlusion, the amplitude of the anacrotic limb of the pulse is scarcely altered. Under lever *3* the immediate effect is the almost complete disappearance of the second steep anacrotic rise, *a;* as the peripheral artery fills this grows but does not regain the amplitude it had during occlusion of the artery. Its beginning also moves away slightly from the top of the anacrotic limb. Under lever *1* the main effect of deocclusion is the disappearance of the steep secondary rise labelled *a*. Presumably, therefore, this wave corresponds with the second steep rise recorded by lever *3*. It is quite clear that it comes considerably later in the pulse cycle in the more central than in the peripheral parts of the artery. It, therefore, seems justifiable to conclude from the evidence presented that this secondary wave is a filling wave running toward the heart from the peripheral artery. It might be added here that shadow records of the same procedure show that the changes in level seen in figure 13 are not due to movements of the artery as a whole, but to differences in fullness.

Shadow records. In these records such up and down motion of the artery as may occur can be recognized and allowed for. The crest line reconstructed from them has essentially the same form (see figs. 9, 10, 11) as has that obtained through the mirror records. A first steep rise, a longer, more gradual rise and a second steep rise, all are present. The main difference is the relative inconspicuousness of the second rise. This difference presumably is to be attributed to the higher peripheral pressures that obtain in the case of the carotid artery, the consequence of which, as has been said, is to increase the diastolic residuum and through that, presumably, the systolic filling, also. Consistent with this explanation is the relatively early appearance of the so-called filling wave, in some of the records at least (in fig. 8, with pulse *5*). In these records the agreement is much closer than in the mirror lever records between the level of the last of the larger steps of the first quick rise on the crest line and the level of the last of the later accelerating steps on the trough line.

In view of the results obtained by both of the methods it seems justifiable to conclude: 1, That during the first steep rise of the crest line the pulse fails to fill the artery quite to its just round size; this stage of decompression probably does not exceed a decrement of 5 mm. Hg. 2, That after the pulse begins to fill the artery and the arterial wall, therefore, to oppose filling, the systolic bore increases relatively little and at an ever decreasing rate, though the rate depends in some measure upon the degree to which the peripheral emptying of the compressed segment is opposed. 3, That later, the interval depending again upon the degree of fullness of the artery beyond the chamber, the resistance which the pulse encounters to its progress beyond the compressed segment leads to its partial and increasing reflection, the effect of which is to cause the systolic caliber of the compressed segment of artery to increase at a much more rapid rate.

RADIAL AMPLITUDE OF OSCILLATION

Mirror levers. The radial amplitude of oscillation is given by the distance between the trough line and the crest line in any given pulse. The variations in radial amplitude can be made out in figures 5, 6 and 7, but in order to present them more clearly they have been plotted separately in figures 14, 15 and 16. The curve of amplitude thus presented rises irregularly and rapidly when the pulse first succeeds in reaching a given lever and turns fairly sharply toward the horizontal at the

moment when, as we have concluded above, the wall of the artery begins to be stretched by the crest of the pulse.

The subsequent course of the curve showing radial amplitude of oscillation differs under the different levers, but upon the whole it is the same under the same lever in the different experiments. The

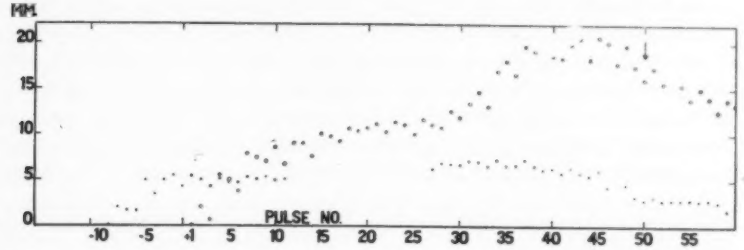


Fig. 14. Radial amplitude of oscillation, dog III. Reconstruction from figure 5.

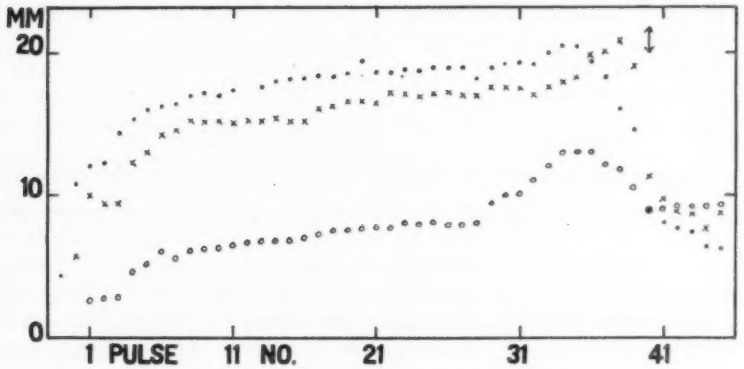


Fig. 15. Radial amplitude of oscillation, dog IV. Reconstruction from figure 6.

curve derived from the records by lever *1* begins to turn slowly downward and becomes quite irregular some time before the fourth sound-phase begins,—some time before the termination of the late accelerating rise of the trough line. A few pulses before the fourth sound-phase begins this descent becomes more rapid but in the vicinity of the first of the fourth sound-phase pulses it turns more or less sharply toward the horizontal.

The radial amplitude curves obtained through levers *2* and *3*, after running their more horizontal course, suddenly turn upward. A tendency to turn downwards, due to the increase in residual blood, may develop before the rise takes place. This inflection invariably occurs earlier under lever *3* than under lever *2;* and, relative to the previous more horizontal part, lever *3* rises further than lever *2*. This rise is due to the appearance of the filling wave described above; it occurs despite the tendency of the accumulation of diastolic residuum to reduce the amplitude. In general, the radial amplitude begins to diminish or ceases to increase before the end of the third sound-phase is reached. Very often the largest diminution in amplitude between any two pulses falls between the last pulse of the third and the first pulse of the fourth

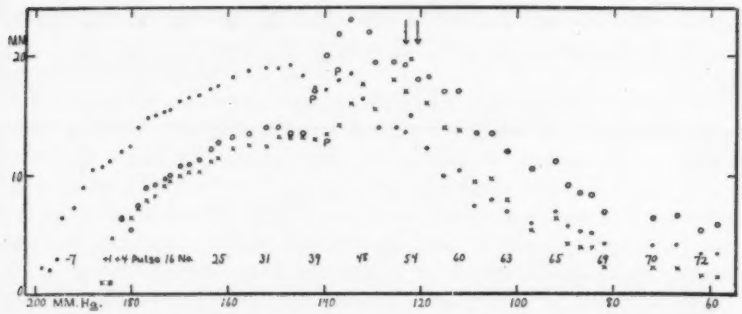


Fig. 16. Radial amplitude of oscillation, dog V. Reconstruction from figure 7.

sound-phase period. In any event the beginning of the fourth sound-phase marks as closely as could be expected the beginning of a curve that tends to become parallel to the base line. This applies to the records written by all of the levers. At times, the initial diminution in amplitude with the beginning of the fourth sound-phase is very marked, and then little further diminution occurs. At other times the diminution is more gradual.

Shadow records. The shadow records, which are taken from only one point on the artery, one coinciding in general with the point upon which lever *3* rests, show essentially similar sequential changes in radial amplitude of oscillation. The variations from record to record, though rather wide, are perhaps no greater than the differences in the conditions might be expected to produce.

Volume oscillations. The amplitude of the oscillations written by a recording sphygmomanometer or given by an oscillometer is determined mainly by the volume of blood entering and leaving the artery within the compression chamber from moment to moment. Although it may at first thought seem strange, it nevertheless is true, that the radial amplitude of oscillation, as we have measured it, sheds but little light upon these volume oscillations. To be sure the radial amplitude of oscillation could be used as the basis of a calculation through which, by taking into account the change in the form of the artery and the diastolic residuum upon which the radial oscillations are built, the area oscillations of the cross section of the artery under a given lever could be roughly determined. In this connection it might be pointed out that for a radial oscillation of given amplitude, the area, and consequently the volume, oscillation will vary as the square of the radial distance of oscillation from the axis of the artery, and that it follows from this relation that in certain stages of decompression the volume oscillation of the artery may actually be increasing while the radial amplitude of oscillation is decreasing. But the maxima of the oscillations of the three levers are not attained at the same moment in any given pulse cycle, so that averaging the areas of oscillation as determined by calculation would be of but little avail in the quest of the pulsatile volume oscillations. For this it would be necessary to have the simultaneous radial amplitude of oscillation of all of the levers from moment to moment. In view of the large experimental errors of the method the results obtained through such a laborious calculation would not justify the time that would be required to make it.

Nevertheless, the radial amplitude of oscillation is of interest through the light it throws upon the factors that enter into the determination of the volume oscillations and upon the play of these factors during decompression.

As is well known, the oscillations of the sphygmomanometer at first slowly increase in amplitude during decompression even at a time when the compressing pressure exceeds that under which the pulse can penetrate into the compressed segment; and later the amplitude more or less abruptly begins to increase more rapidly. The present records throw no light upon the mechanism of the first of these two stages; they do however account for the second. It is due both to the increasing depth of penetration of the pulse, as have been pointed out, and to the growth of the more proximal radial amplitude of the oscillations. The increase in the oscillations of the sphygmomanometer then progresses

more slowly again. The behavior of the radial amplitude of oscillations indicates that this is to be expected.

In many of the records the variations in radial amplitude indicate that there might be a tendency for the volume oscillations to show two maxima, one at the time the diastolic residuum is rapidly accumulating and before the filling wave develops, and one while the filling wave is in evidence. It is, therefore, of interest to note that the sphygmomanometric records obtained from man do sometimes display two maxima (1).

It is evident that the diastolic form of the artery at the time the volume oscillations are maximal is not necessarily always the same. As has been said, it was at one time supposed that maximal oscillations coincided with diastolic compression and that at this time the artery collapsed practically completely during diastole. MacWilliam and Melvin (9) maintain that maximal oscillations do not develop until the flattening of the artery reaches such a degree that the shorter diameter of the tube is (roughly) something like half that of the passive circular tube. Judging from the radial amplitude of the oscillation as presented by our records it seems fair to conclude that the form of the artery in diastole at the time the largest volume oscillations are in evidence, while never completely flat, will depend upon a number of conditions, such as duration of pulse cycle, resistance to outflow, etc., etc., that vary from estimation to estimation, some of which are not the same in different parts of the compressed artery. The degree of flattening at the time maximal oscillations are recording is very variable.

When determining the pressure by the method of continuous escapement, the amplitude of oscillation at a certain stage "will diminish more or less abruptly." The pressure indicated by the sphygmomanometer at this moment "has been advocated as the index to the minimum (diastolic) pressure" (1). MacWilliam and Melvin (9) regard the level of external pressure just *after* the abrupt diminution in amplitude has taken place as a better guide to the diastolic pressure. Judging by the behavior of the radial amplitude of oscillation it seems justifiable to conclude that the diastolic pressure ordinarily will lie between these two points in the position where the accelerating decrease in amplitude of the oscillations of the sphygmomanometer changes to a retarding decrease.

THE PRE-ANACROTIC PHASE

Mirror records. a. The pre-anacrotic waves. The pre-anacrotic waves with which this section deals are those that first appear with or close to the beginning of the critical period of decompression, the period included between beginning penetration of the pulse and the beginning of the fourth sound-phase. Their consideration can best be begun by describing in turn the metamorphosis they pass through under each lever during decompression. For reasons which will soon become obvious, the pre-anacrotic waves as they appear under lever *3* will be described first.

Lever 3. Dog. III. The lever is quite motionless up to pulse *−5*. With pulse *−5* the long, low negative wave, which has already been referred to as the gravity wave, begins to develop. Its beginning in any cycle is exactly synchronous with the beginning of the pulse as written by lever *1*. By pulse *−2* this wave has become quite evident. With pulse *1*, better with pulse *2*, the phase of the pulse corresponding in time with the negative wave can be seen to be made up of two parts. The first part is a rather indefinite depression beginning, as before, exactly at the same time as the same pulse as written by lever *1;* the second consists of a notch-like depression separated from the preceding depression by an indistinct positive wave. Furthermore, the second negative wave is much later than the pulse under lever *1*, and its ascending phase passes directly into the anacrotic limb of the pulse. When this particular pre-anacrotic wave is followed through the record it is found that it steadily grows briefer, at the same time that it increases in depth. This deepening is rapid at first, until, with pulse *4* or *5*, a depth of 1.5 mm. is attained. It then grows more slowly to a depth of about 3 mm. in pulse *42*, when it rapidly deepens to reach 4 mm. in pulse *49*. With pulse *50*, the pulse in which the signal of the change to the fourth sound-phase falls, it begins to rapidly diminish in depth and it has practically disappeared in pulse *55*. While this wave thus grows, changes take place immediately in advance of it. With pulse *16*, possibly earlier, the disturbance that precedes this notch dissociates into two very much finer waves of low amplitude, all eventually forming a complete series of waves terminating in *the* very deep pre-anacrotic negative wave. All become briefer as decompression proceeds. These preliminary pre-anacrotic waves disappear completely from view at about the time (pulse *50*) *the* pre-anacrotic wave begins to diminish

in depth. It was with pulse *52* that the accumulation of the diastolic residuum reached its maximum acceleration.

Dog. IV (fig. 3). In pulse *−1* the gravity wave, the long, low negative wave synchronous with the positive pulse written by lever *1*, is clearly seen. In pulse *1* it is shortened by the appearance of a relatively short and sharp negative wave from the ascending limb of which rises the anacrotic limb of the first real pulse. Again, with decompression, this pre-anacrotic notch increases in depth and in brevity, presumably much in the same way as in the case of dog III, but, owing to certain changes which will become clear in a moment, the measurement of depth does not in this case give a clear conception of the growth of the wave. And again, preliminary pre-anacrotic waves soon appear (with pulse *4*, possibly earlier), and then grow into a perfect series of three crescendo waves of which, as may be seen in pulse *14*, *the* pre-anacrotic wave is the deepest and is preceded by a very distinct positive wave. With further decompression the trough of the last pre-anacrotic wave begins to rise and eventually, as a result of this process, it comes to form a part of the anacrotic limb, proper, of the pulse. In the meanwhile the wave next preceding it has so grown in depth and has so changed in other respects that it now (pulse *33*) resembles and takes the place of the wave that has passed into the anacrotic limb. The depth of all of these waves increases to pulse *36* or *37*. It diminishes, however, in pulses *38* and *39;* and in pulse *40*, the pulse in which the fourth sound-phase begins, every trace of the pre-anacrotic waves suddenly disappears. It was with this pulse that the acceleration of the accumulation of the diastolic residuum stopped, and that the terminal dip of diastolic phase of the pulse became less steep.

Dog. V (fig. 4). In this case lever *3* is motionless down to pulse *−3*, possibly to pulse *−4* (about 188 mm. Hg.); here the lever is depressed very slightly to form a long, low negative, or gravity, wave. This wave is seen again with increasing distinctness in pulses *−2* and *−1*. With pulse *1* there is a sudden change. The long, low negative wave disappears or is replaced by a low positive wave and this is followed by a relatively deep and sharp negative wave, or notch, the ascending limb of which is continued into what may be regarded as a rudimentary anacrotic limb of the pulse wave. The low preliminary positive wave preceding the notch soon comes to be replaced by a series of indistinct periodic vibrations, of which more in a moment. Tracing the notch through the record, it at first becomes briefer, as do all of the other initial processes of the pulse. Soon, however, it becomes apparent that the increas-

ing brevity of the deep negative wave is in part due to the encroachment upon it of the anacrotic limb of the after-following pulse. This not alone makes the negative wave briefer, but in time shallower also (pulse *29*), and eventually the remnant of this negative wave comes to be lost in the anacrotic limb of the pulse (pulse *45*), which now rises sharply as a part of the ascending limb of a preceding preliminary wave, the origin of which may now be traced.

As the vibrations that come to replace the slight rise preceding the negative wave, where it first develops, grow in distinctness it becomes apparent that they are a crescendo of periodic vibrations, and that the wave that has just been traced as *the* negative wave is really one of this series. The whole series, at least at first, is built up on a sharp decline—possibly a gravity depression—from the bottom of which the anacrotic limb of the pulse starts. The depth of whichever of the negative waves as happens to be deepest, at first increases slowly, then, beginning with pulse *44*, more rapidly. The wave diminishes slightly in depth in pulse *52;* and practically disappears in pulse *53*, the pulse with which the fourth sound-phase begins, returns and attains its maximum depth of 4.5 mm. with pulse *54*. With the next pulse, *55*, however, it disappears completely and definitively, and there is left of the preliminary vibrations only a vestige of one of the positive waves. Again, it was with these very pulses that the acceleration of the rise in the diastolic residuum came to an end.

Lever 2. We shall next describe the pre-anacrotic waves that develop in the middle region of the compressed segment.

In the record of *dog III* the presence of long waves, presumably of respiratory origin, coupled with some peculiarity in the form of the pulse and a lack of definition, makes the tracing of the preliminary waves somewhat uncertain. A long, low negative wave seems to be present in pulse -8. In pulse -5 this is shortened by the appearance of the anacrotic limb of the pulse wave. It becomes briefer and possibly deeper for a few pulses. It is still evident, but as a relatively shallow wave, in pulse *10* where the record temporarily becomes so indistinct that it cannot be read. In the vicinity of pulse *35* the record becomes clear again, and here can be seen a negative wave, about 2.5 mm. deep, from the bottom of which ascends the anacrotic limb of the pulse. The subsequent fate of this negative wave cannot be followed further in this record. But in another record made immediately afterwards from the same animal the pre-anacrotic wave seems to disappear a pulse or two before it disappears in the record written by

lever *3*. At no time can anything more than a single, simple negative wave be discerned in the pre-anacrotic region. At all times the beginning of this wave is exactly or nearly synchronous with the pulse recorded by lever *1*.

Dog IV. The record chosen for reproduction (fig. 3) does not begin at a sufficiently high compressing pressure to show the early development of the pre-anacrotic waves. Its description is therefore pieced out by that of a record obtained immediately after it from the same animal, but which does not lend itself so well to reproduction as a whole as does the one selected for that purpose.

A very distinct preliminary pulsation can be made out under the highest compressing pressures. When it takes sufficiently definite form this movement seems to follow very closely the configuration of the pulse written in the same cycle by lever *1* and consists of two very low positive waves with a very indistinct depression between them. With pulse -5 (the one in which the anacrotic limb appears under lever *1*) the depression between the two indistinct positive waves becomes the most marked feature of the pulse and eliminates the second positive wave. In point of time the first positive wave is practically synchronous with the first positive wave of lever *1*, while the descending phase of the negative wave of lever *2* corresponds with the ascending phase of the main pulse written by lever *1*. With pulse *1* this negative wave has already attained a depth of 3 mm. Its beginning here and throughout the record is exactly synchronous with the pre-anacrotic limb as written by lever *1*. Presumably it is the gravity wave. With pulse *15* it can be seen that this negative wave is made up of two parts; the first part is slow and its beginning remains synchronous with the beginning of the pulse as written by lever *1;* the second part, therefore, begins later than the pulse under lever *1*. In general this second wave at first increases rather rapidly in prominence and then maintains itself without any material change until, again with pulse *40*, it suddenly disappears.

Dog V. Under the high compressing pressure lever *2* records with each pulse a couple of waves so low as almost to escape detection. These waves become even less distinct as the critical compressing pressure is approached (pulses -8 to -6). This is the only evidence to be seen here of a long, low gravity wave, and it is circumstantial only. In pulse -5 practically all that can be made out is a minute notch appearing, in point of time, a bit later than the crest of the same pulse as recorded by lever *1*. The notch increases in prominence to pulse

−2. Here the pulse proper appears under this lever, and its anacrotic limb includes the ascending limb of the notch. The notch is distinctly later than the pulse under lever *1*. The notch now diminishes in depth and practically disappears, the pre-anacrotic phase appearing to be occupied in its place by two or three indistinct and very fine waves. These fine waves become somewhat more distinct and increase somewhat in amplitude with further decompression but suddenly disappear in pulse *50*, the third pulse from the last of the third sound-phase period.

Lever 1. We have yet to describe the pre-anacrotic region under the most central of the three levers.

In *dog III*, under the highest compressing pressures, lever *1* records a long, low positive wave. When the pulse begins to penetrate to this lever the anacrotic limb rises out of this preliminary wave. What remains of the preliminary wave therefore becomes a pre-anacrotic positive wave. Vestiges of this wave can be traced through only a few pulses. A pre-anacrotic negative wave is at no time evident.

Dog IV. Two long, low positive waves are recorded under the high compressing pressures. With decompression a slight notch develops between them (pulse *−10*). The first of the two waves shortens materially with decompression. With pulse *−4*, possibly with pulse *−5*, the second of the two increases very decidedly in amplitude; this is the mark of the penetration of the pulse to this lever. The notch between the two waves is most marked in pulses *−5*, *−4*, *−3* and *−2*. With further decompression it soon (pulse *10*) fails any longer to pass below the general level of the record, the pulsatile movement then consisting of the first low positive wave, which now has become quite brief, separated by a depression, though not a negative wave, from the after-coming pulse proper. Later, however, (somewhere between pulses *15* and *20*) the depression again is converted into a short, sharp negative wave. This negative wave increases gradually in depth to a maximum of about 1.5 mm., then remains constant through pulse *31*, after which it decreases in depth to practically disappear in pulse *36*. The short pre-anacrotic positive wave in the meanwhile becomes very brief and inconspicuous and disappears in pulse *28*.

Dog V. The preliminary wave recorded in the upper parts of the compressed artery consists in this case of a single, long-drawn-out positive wave. Evidence of the penetration of the pulse to this lever is first seen in pulse *−12*, where a distinct peak makes its appearance on the crest of the preliminary wave. With decompression, the pulse proper advances into the preliminary wave which now is a pre-ana-

crotic wave. Vestiges of this pre-anacrotic positive wave can be traced as far as pulse *45* or thereabout, though it early becomes a very insignificant feature. A negative pre-anacrotic wave is never visible in the record of this lever.

b. The anacrotic limb. Certain changes that occur in the foot, if not in the whole of the anacrotic limb of the pulse are related to the pre-anacrotic waves and therefore should be considered in connection with them. These changes are fundamentally alike in all of the records, but they occur in different varieties. Reference has already been made to two of them, namely, the passage of pre-anacrotic waves up into the anacrotic limb, and the addition to the anacrotic limb of what we have called the filling wave. In general it may be said (see figs. 3 and 4) that when the pulse first succeeds in making its way through the compressed artery the anacrotic limb starts gradually from the end of the diastolic phase and rises in relatively gentle curves. Later, however, its start becomes very abrupt and the gradient straight and steep. In general the initial gradient increases as the pulse passes along the compressed segment. This becomes obvious when in any one pulse a comparison is made of the thickness of the foot of the anacrotic limb under the three levers. Measurement of the record also brings it out clearly. By way of example, the actual time consumed by the lever in moving from the beginning to the end of the straight anacrotic rise in a typical pulse may be cited. In pulse *30* of figure 3 this time for levers *1*, *2* and *3* is 0.032, 0.011 and 0.007 second, respectively. The abruptness of the rise may be so great as to actually throw lever *3*—sometimes lever *2* also—from the artery. This happens to be the case in record reproduced as figure 3, in which vibrations at the top of the anacrotic limb, undoubtedly due to fling of the lever, are to be seen beginning under lever *3* in pulse *5*, under lever *2* in pulse *25*, while we cannot be certain that lever *1* is at any time thrown from the artery. In the case of lever *3* a sudden diminution in the gradient of the foot of the limb takes place in the very pulse in which the pre-anacrotic waves disappear. This often occurs under levers *2* and *1* also, but usually it takes place earlier under lever *1* than under levers *2* and *3*, and sometimes earlier under lever *2* than under lever *3*. The sudden diminution in the abruptness of the rise of the anacrotic limb under lever *3* in the very pulse in which the pre-anacrotic waves disappear is a very striking phenomenon.

Summary. The pre-anacrotic waves that remain after disregarding the so-called preliminary and gravity waves, together with the very

abrupt ascent of the foot of the anacrotic limb, we now designate the *pre-anacrotic phenomenon*. The form this phenomenon takes, at first sight so complex and varied, is in reality comparatively simple when a composite is made of the varieties, which may now be attempted.

Under *lever 3*. When the pre-anacrotic phenomenon first appears under the distal lever it usually is composed of a single relatively long negative wave, but later it becomes briefer and its form may be modified by the appearance of a series of smaller waves in front of it and by the encroachment upon it of the anacrotic limb of the pulse from behind, as a result of which the last of the series of vibrations may pass up into the anacrotic limb. The first signs of the phenomenon become manifest only after the record gives evidence that the artery contains a residuum, though it may be only slight, of blood, and with the arrival of that pulse at the position of the lever which is of sufficient volume to produce a distinct anacrotic limb. It disappears, usually with the greatest of abruptness, with the first pulse of the fourth sound-phase.

In one of the three animals (*dog III*) the pre-anacrotic phenomenon disappeared gradually. This happened to be the animal in which the sounds were not well heard. In the case of this animal the signal indicating the beginning of the fourth sound-phase in every record fell amongst the pulses in which the pre-anacrotic negative wave was rapidly diminishing in depth; it was never more than four pulses removed from the one in which the negative wave definitively disappeared.

Under *lever 2* the pre-anacrotic phenomenon is in evidence during the whole of the critical period of decompression in the case of one animal only (dog IV); that is to say, this is the only case in which it is in evidence from the time the artery begins to hold residual blood and until the fourth sound-phase begins. In other cases it may be present at the beginning of the critical stage, but usually soon disappears.

Lever 1. The pre-anacrotic phenomenon in the form of a negative wave has been seen under lever *1* in but one animal (dog IV) and in that one it appeared in typical form only during a part of the critical stage of decompression.

The slower depression at the close of diastole which has been designated the gravity phenomenon, as has been pointed out in an earlier section, in any one pulse begins at the same instant under all of the levers; it is not a propagated phenomenon. On the other hand the pre-anacrotic phenomenon unquestionably is propagated, travelling at approximately the same rate as the pulse. It is upon this difference, in addition to others to be considered below, that we base the distinction we have drawn between them.

The movements of the levers determined by the pre-anacrotic phenomenon are very close to the limit of their capability; consequently it is quite possible that the records do not reproduce it in its true form. But if there is any distortion, it cannot be very great, because of the similarity between the lever and shadow records of the phenomenon to be now considered.

Shadow records. The shadow records show the movements of the artery as they occur at one point only, namely, the one corresponding approximately with the position occupied by lever *3;* consequently, they throw no light on the propagation of the pre-anacrotic phenomenon. But the shadow records furnish the basis for distinguishing between translational movements of the artery and wave movements of its fluid contents. Translational movements of the artery are indicated by movements of the two sides of the shadow in the same direction, while wave movements in the blood, like the movements caused by the pulse itself, cause the opposite walls to move in opposite directions, and the extent of the movement on the two sides should be the same, excepting such modification as may occur through the unequal action of gravity and through such accidents as unequal drying of the artery. The only other motion of the artery that would cause the two edges of the shadow to move in opposite directions would be rotational movements of the artery while flattened. It would occupy too much space to present here all of the reasons that have served to convince us that if rotational movements are a factor at all in producing the pictures now under consideration, they certainly are negligible. We can only say now that the close agreement of the results obtained through the mirror levers, especially from the models to be described subsequently in which a movement of rotation is impossible, with those obtained by means of the shadow records leaves no room for doubting the wave nature of the pre-anacrotic phenomenon.

It has been seen that some of the shadow records show what we take to be gravitational depression of the artery in the pre-systolic phase, especially, early in the critical period of decompression. In addition, all of the shadow records clearly exhibit movements corresponding in position and character with those of the pre-anacrotic phenomenon as recorded by the mirror levers. These are readily distinguishable from the gravity depression. It would take too much space to describe in detail the character and metamorphosis of the pre-anacrotic waves in each of the records; for, as in the case of the mirror records, the form taken by the phenomenon is extremely varied. It must suffice to refer

the reader to the one complete record reproduced here as figure 8. Here can be seen, *a*, the short, sharp waves, the last of which is negative and terminates in the anacrotic limb of the pulse; *b*, the appearance of these waves during decompression shortly after a diastolic residuum of blood remains in the artery; *c*, their growth for a time in amplitude and their increasing brevity; and *d*, finally their more or less sudden diminution and eventual disappearance, within the limit of error, with the termination of the accelerating and irregular rise of the trough line.

The shadow records, though, are especially valuable in showing that while the amplitude of the pre-anacrotic phenomenon on two sides of the shadow may be very different, the waves on the two sides in any given pulse cycle are the same and, allowing for the gravity depression, they invariably are in the same phase with respect to the axis of the artery. This constitutes the proof that the pre-anacrotic phenomenon is due to wave motion of the blood and not to motion of the artery as a whole.

We reproduce a few enlarged pulses from another of these records (dog VII, fig. 17) in order to indicate not alone that we are not dealing with translational movements but also the variety in which the phenomenon presents itself, and the remarkable brevity and relatively great amplitude the waves may have. This record, it should be recalled, was obtained from an artery that had dried somewhat on its upper surface.

THE PRE-ANACROTIC PHENOMENON IN MODELS

Several types of models have been used in an effort to arrive at an understanding of the pre-anacrotic phenomenon as seen in the artery *in situ*. As the results obtained with all types are essentially alike we shall describe in detail only the simplest. It is one that can easily be made by anyone interested. The inner tube of a bicycle tire, opened by cutting out the segment carrying the tubulation, is closed at one end by a perforated cork of the same circumference as the tire. Through the perforation the interior of the tire is in communication with a pressure bottle. To the other end a piece of heavy-walled, but flexible, rubber tubing is joined by means of a short glass tube, both of the same circumference, approximately, as the tire. The other end of the heavy-walled tube ends blindly. The system is filled with water to the total exclusion of air and lies horizontally on a table so that, as the tire empties, it flattens in the horizontal plane. The pulse is generated by compressing the heavy-walled tube with the hand. The length of the

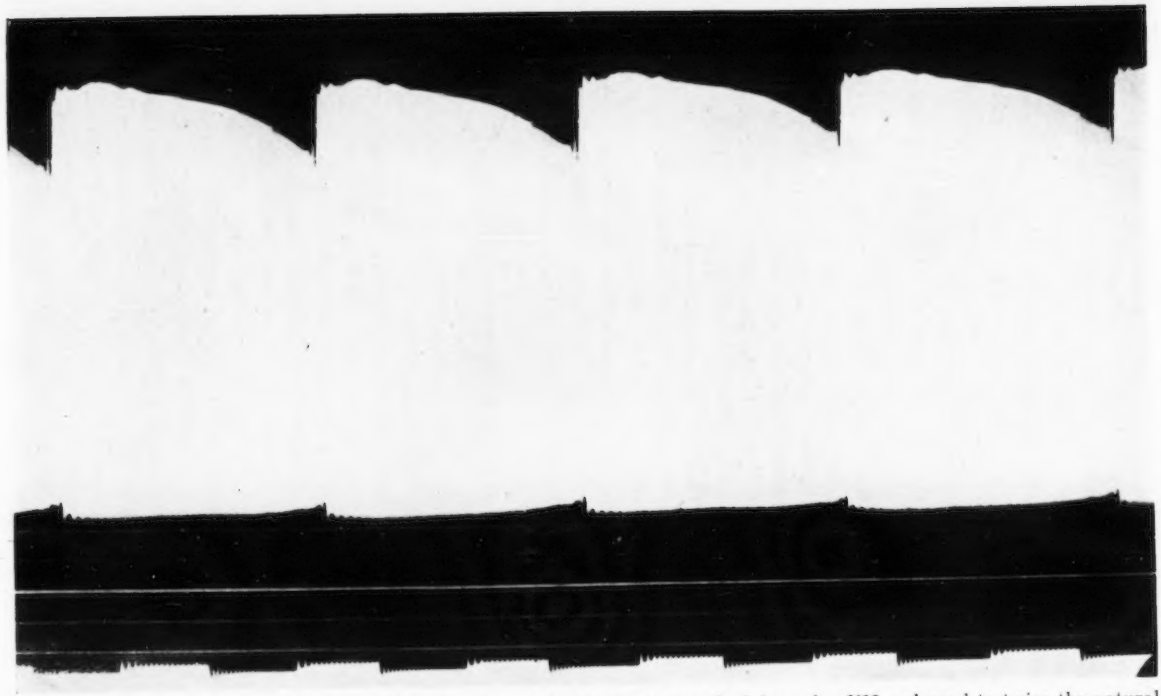


Fig. 17. Five pulses from a decompression record made by the shadow method from dog VII, enlarged to twice the natural size, to illustrate one of the several varieties of the pre-anacrotic phenomenon. The time is indicated in fifths of seconds.

system is such, up to 1 meter, that waves reflected from the far end can readily be discounted. A system of smaller bore, the tube of which was made of rubber dam, was first employed, and most of our observations have been derived from it.

Records have been made both by means of levers writing on smoked paper, and optically. The levers were very light and magnified the movements only about four to five times. They rested horizontally across the "artery" and were fastened to it by means of vaseline. The optical method of making records was patterned after the one employed in obtaining the records from the artery in situ. Parallel rays were thrown horizontally across the "artery," which lay just behind two vertical slits. The upper edge of the "artery" behind the slits was focussed onto a photoregistering apparatus with aperture set vertically. The image of one slit fell directly upon the aperture, while that of the other, by means of a prism, was brought on to it just above the former. The magnification was $\times$ 2.8. The time was recorded usually in hundredths of seconds. The results obtained by both the mechanical and the optical methods of registration are essentially alike.

Lever records. The results obtained vary with the fullness of the "artery." We begin by describing the pre-anacrotic phenomenon as presented by the rubber dam "artery" filled to a depth of 8 mm. As the just round diameter of the "artery" was 14 mm., the condition of the tube in this experiment may be described as partly flat. The pressure within the "artery" is practically zero against the upper wall and something over 8 mm. H_2O against the lower wall. The "artery" at the two ends under these circumstances has the form of a flattened conoid, but quickly takes on the form of a cylindroid flattened below, provided, as in the present case, it contains sufficient water to bulge the upper wall above the lateral ridges that otherwise tend to form where the rubber is folded on itself. The distance down the tube at which the cylindroid becomes practically uniform varies somewhat with the state of fullness of the "artery" and with the longitudinal tension it is under.

Two levers are used. They are attached rigidly to a single stand in such a way that they cross the "artery" exactly 1 cm. apart. The lever writing the upper of the two tracings (fig. 18) is the more central of the two. The first record is obtained (two pulses, or more if necessary to secure uniformity) from a point close to the central end of the "artery" and for the next record the levers are moved along the "artery" a distance of 1 cm. so that the more central lever now occupies the posi-

tion the distal lever occupied for the preceding record. Only when the levers that occupied the same position on the "artery" in successive shifts record identical pulses can it be said that the successive pulses were the same, and that the successive records, therefore, are comparable. The levers were thus shifted in the experiment illustrated by the figure until records were obtained 22 cm. down the artery. In a completing experiment, with the drum revolving at a somewhat slower speed, records were made with a single lever placed at greater intervals along the length of the "artery" down almost to its end. The combined experiment required 54 sets of curves for the complete series. A complete picture of what transpires along the course of the "artery" can be obtained alone by studying all of the records of the series. Unfortunately, however, they cannot all be reproduced here. But some idea can be conveyed of the changes occurring during propagation of the pulse by reproducing selected curves and filling in the gaps by text descriptions.

In the figure (fig. 18) it is seen that at 3 and 4 cm. the inclination of the anacrotic limb of the pulse is relatively slight and its beginning quite gradual. At 9 and 10 cm. the anacrotic limb is much steeper and its start very much more abrupt. The change to this form develops gradually between the proximal end of the "artery" and the 10 cm. position. At 11 cm. a pre-anacrotic negative wave begins to appear. At 12 cm. this wave is somewhat deeper and longer, and still deeper but briefer at 14 cm. At 15 cm. it comes to be preceded by a small positive wave. At 17 cm. the negative wave does not dip so deep below the base line as it did, while the positive wave ahead of it has grown and is preceded by the beginnings of another negative wave. At 18 cm. these changes have progressed further. At 19 cm. the first negative wave and the positive wave have become parts of the anacrotic limb which now rises out of a negative wave that has come into existence much in the same way as did the first negative wave. Continuing the description by using the completing series, and allowing for the slower rate of motion of the paper, it is seen that at 28 cm. the curve again has practically the form it had at 15 cm.—the anacrotic limb is preceded by a deep negative wave and this in turn by a small positive wave. Well up on the anacrotic limb a slight irregularity is indistinctly to be made out; this, we think, is the remnant of the negative wave that was seen moving up the anacrotic limb at 19 cm. At 30 cm. the second negative wave and practically the whole of the positive wave preceding it have been taken up into the anacrotic limb, in the upper third of

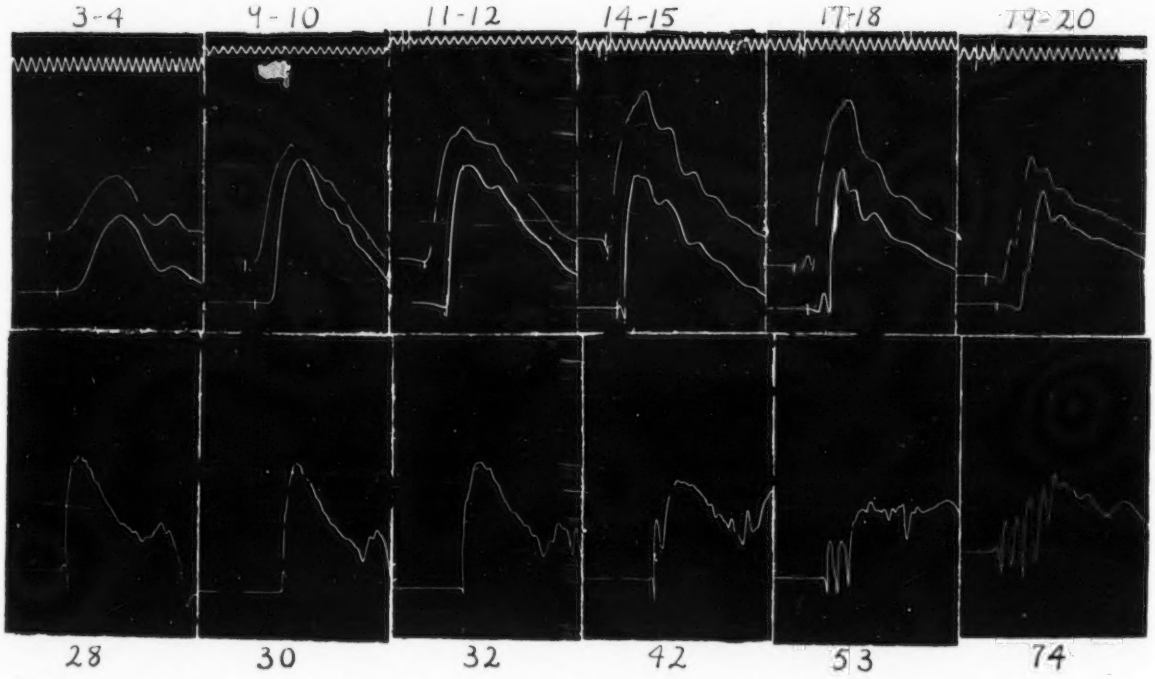


Fig. 18. Lever records from the rubber dam "artery." The numbers give, in cm., the distances down the "artery" of the point from which the corresponding records were taken. Where two numbers are given the first refers to the upper of the two curves. The tuning fork makes 100 d. v. per second.

which remnants of the first series of waves can still be seen. Still another negative wave begins to form before the anacrotic limb at the 32 cm. position, and here two breaks on the anacrotic limb presumably indicate the positions the two engulfed negative waves now occupy. At 42 cm. the pre-anacrotic waves have developed to the stage that the preceding series of pre-anacrotic waves had at 28 cm. and at 15 cm. Deep notches are now seen in the anacrotic limb; these probably are the small waves, which have passed into the main wave, metamorphosed by some process so as to assume this prominence. As these waves grow, one after another, in front of the pulse, they continue to move on in advance of the pulse, growing larger and larger as they proceed. Thus at 53 cm. we see two fully developed, large negative waves separated by positive waves and a third negative wave developing; and at 74 cm. four fully developed negative waves, separated by positive waves and a fifth negative wave in a more advanced stage of development than the developing wave seen in the 53 cm. position.

Shadow records. Entirely comparable records have been obtained from the tire model by both the lever and the optical methods. We shall present the optical records of one such experiment. In this case the tire was filled to a depth of 1.5 cm.; consequently the tube again was partly flat. The experiment was done in two stages. In the first, the distance between the two slits was 3.2 cm.; in the second the lacunae in the more important stages of the first part were filled in by records made with the slits 1 cm. apart. The beam of light was shifted along the length of the "artery" for the successive records, just as in the case of the levers, in order to control pulse form. Excepting accidental movements, the records selected from the continuous series for purposes of illustration (fig. 19) show, as the wave progresses down the artery, (*A* to *B*) the increase in the abruptness of the wave front; (*C* and *C'*) the appearance of a small negative wave; (*D* and *D'*) the appearance and growth, between this and the anacrotic limb, of a sharp positive wave, and of a negative wave between the latter and the anacrotic limb (*E* and *E'*) the deepening of the notch between the sharp positive wave and the anacrotic limb until it comes to form a deep, sharp negative wave from the bottom of which the very steep anacrotic limb ascends; (*F* to *H*) the ascent of these two sharp waves into the anacrotic limb and (*G* to *I'*) the formation of a similar series of waves in front of the ascended waves; and finally (*J*), after the pulse has progressed a considerable distance along the tube, the tendency for the anacrotic limb to break up into a series of sinuous waves.

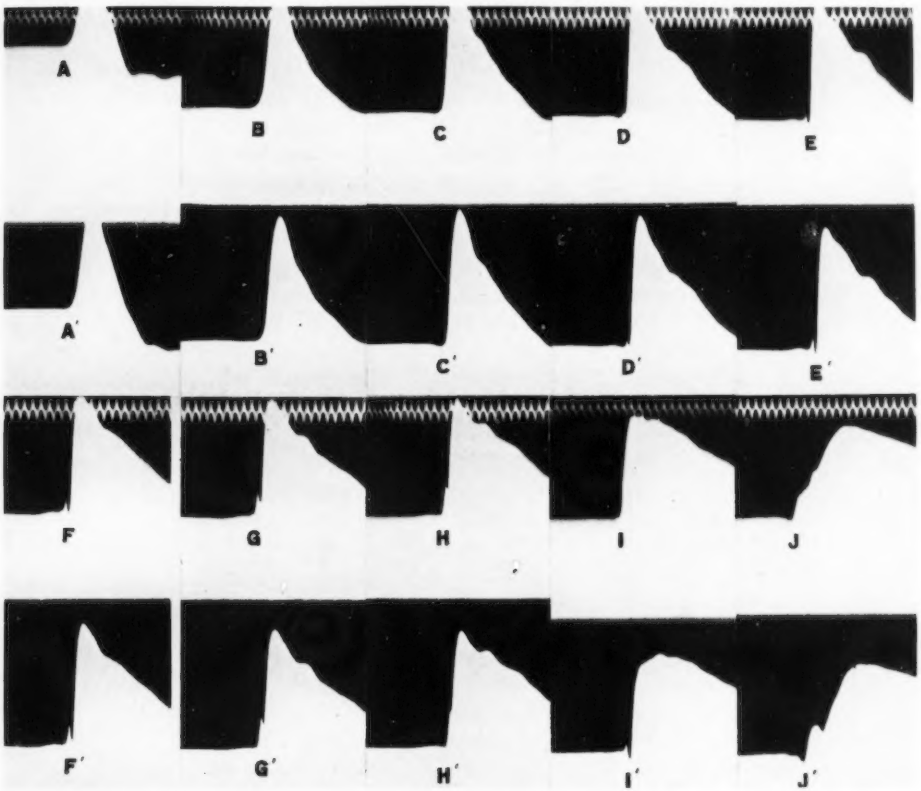


Fig. 19. Shadow records (reduced) showing the development of the pre-anaerotic phenomenon with the progress of the pulse along the rubber tire "artery." The "artery" is partly flat. The tuning fork makes 100 d. v. per second. The wave front is to the left. The distance in cm. the pulse has progressed along the "artery" is: $A = 5.8$, $A' = 9.0$; $B = 16$, $B' = 17$; $C = 18$, $C' = 19$; $D = 20$, $D' = 21$; $E = 23$, $E' = 24$; $F = 26$, $F' = 27$; $G = 28$, $G' = 29$; $H = 30$, $H' = 31$; $I = 34.6$, $I' = 37.8$; $J = 50.6$, $J' = 53.8$.

Experiments upon the nature of the pre-anacrotic phase and conditions modifying it. These experiments are still in the preliminary stage. In the first place we call attention to the fact that the distance down the "artery" at which the pre-anacrotic phenomenon makes its appearance varies with the fullness of the "artery." Exact determinations of this relation have not been made. The results of a rough determination in the case of the rubber dam "artery" are given in table 5. In this case the internal pressure at which the distance becomes infinite, i.e., at which the phenomenon disappears, amounts to only about 4 mm. Hg., which is about the pressure necessary to give to the tube of 14 mm. a cylindrical form. Qualitative observations on the tire model reveal the same shifting of the phenomenon as the tube fills and empties.

The phenomenon, naturally, can not develop when the pulse is made to traverse the completely empty "artery," and the negative waves of

TABLE 5

DEPTH OF LAYER OF FLUID IN ARTERY	PRESSURE NECESSARY TO GIVE THIS DEPTH	PREANACROTIC PHENOMENON FIRST APPEARS
mm.	*mm.* H_2O	*cm.*
2.0—	2	7±
8.0	8	11±
10.0	22	12±
11.0	32	13±
11.5	44	33±
12.0—	60	∞(?)

the phenomenon increase in depth for a time as the thickness of the layer of the liquid in the "artery" increases. Figure 20 illustrates these statements. Here a series of pulses is sent down the "artery" as it gradually fills from the completely flat position. The layer was 0.8 mm. deep when the phenomenon became evident.

The phenomenon in models is not due to flattening of the "artery" against the support upon which it rests; for it does not disappear in the rubber "artery" suspended by horizontal traction under water (see below). Neither is it produced by the elastic properties of the conoid through which the pulse travels to reach the flattened "artery;" for replacing this with a metal tube shaped like the cone, and tapered so that its distal orifice has exactly the shape of the partly flattened "artery" into which it is inserted, does not alter the phenomenon in any essential respect. Nor is it due to the asymmetry of the beginning

of the tube produced by its sag onto the table. This sag can be removed by supporting the tire between two stretched sheets of rubber dam, one below, the other above the tire. The "artery" thus supported still displays the pre-anacrotic phenomenon.

Fig. 20. Effect of increased filling of the "artery" on the depth of the negative wave of the pre-anacrotic phenomenon. The artery is completely flat at the start.

Fig. 21. Shadow record showing the pre-anacrotic phenomenon on the upper apex, *a*; the lower apex, *b*, and the lateral edge, *c*, of the partly filled bicycle tire "artery" stretched horizontally under water. The pulse front is to the left. The tuning fork makes 100 d. v. per second. Reduced to half size.

By suspending the bicycle tire "artery" horizontally under water in a trough with glass walls it has become possible to obtain records of the movements of the upper and lower apices by the shadow method as has been done with the artery of the animal in situ. The records so obtained (see fig. 21) agree with those obtained from the artery in showing that the pre-anacrotic waves are due neither to wave movements of the "artery" *en masse* nor to translational movements. If they were, the waves on the upper and lower wall of the partially flattened tube would parallel each other. As a matter of fact they are almost, though, for some reason that is not yet clear, not quite, in the same phases with respect to the axis of the "artery." Simultaneous records of the displacement of the tube wall at its upper and lower apices and at its lateral edge in the circumference of a given plane of section at right angles to the length shows (fig. 21), as might have been anticipated, that in general when the pre-anacrotic waves cause outward motion of the upper and lower apices, inward motion of the lateral edge occurs simultaneously.

It is clear from these experiments that a rubber tube will display the pre-anacrotic phenomenon only when it is filled short of complete flattening and up to a slight positive pressure, not more than 4 and 5 mm. Hg., under which it becomes just about round.

That the cause of the disappearance of the phenomenon at the latter grade of fullness of the "artery" is not the pressure per se but rather the assumption of the cylindrical or nearly cylindrical form under the pressure from within is readily demonstrable. For if the internal pressure be raised to the point at which the phenomenon completely disappears, and the proximal third of the "artery" be then strapped down with rubber dam in such a way as to restore the flattened shape in the proximal end of the tube the phenomenon reappears.

The experiments thus far done do not suffice to determine the nature of the pre-anacrotic phenomenon. Indeed, the problem in its further development becomes one in mathematical physics; and it would be futile to attack it theoretically until more is known quantitatively with regard to the fundamental mechanical characteristics of the system with which we are dealing. It is possible that the motion is related, at least in some respects, to the so-called deformation vibrations occurring in drops and jets of fluids, first described by Lenard (13) and worked out mathematically by Lord Rayleigh (14). At present though, we are inclined to account for it upon the basis of another manifestation of surface properties, also worked out by Lord Rayleigh, namely the ripple phenomenon (15). We are not as yet prepared to say definitely how the pre-anacrotic waves form, but nevertheless venture the following hypothesis, which it should be added, differs in some respects from the one offered by Lord Rayleigh for ripples.

As the pulse advances along the artery it forcibly pulls its walls apart. As long as the tube is partly flat a rather sharp angle is thus formed in the wall of the artery with its apex located at the point where the head of the pulse carries the wall above its partially collapsed position. This angle increases the tension of the wall, and this tension tends to eliminate the angle. But it can be eliminated only by the displacement toward the angle of fluid from the artery ahead of and behind the anacrotic limb of the pulse. Thus, as the pulse proceeds, a negative wave eventually develops ahead of it, and the gradient of the main wave steepens. But now the negative wave also produces an angular deformation of the artery in advance of it. This angle, however, is so directed as to force fluid away from it, with the result that a positive wave begins to form ahead of the negative wave, and so on.

The passage of the pre-anacrotic waves up into the anacrotic limb, which can be so clearly traced in the records just described, early stages of which are also seen in the records obtained from animals, is

probably due in part to the fact that the pulse is large and the ripples small; and in part to the fact that the main wave, if ample enough, or (and) if the artery is full enough, distends the artery, while the ripples do not. For, as is well known, large waves travel faster than small waves; furthermore, waves that are confined by the elasticity of the walls of the tube will travel faster than the smaller waves which, practically, are propagated by gravity alone. The result is that the primary wave overtakes and engulfs the ripples. If this is the correct explanation of the over-running of the ripples by the pulse, it would follow that a pulse that does not distend the artery would not overtake the ripples, at least, not so quickly. This actually is the case.

THE DISAPPEARANCE OF THE PRE-ANACROTIC PHENOMENON AS AN INDEX TO THE DIASTOLIC PRESSURE

But, whatever the nature of the pre-anacrotic phenomenon, the fact remains that it is in evidence in tubes only when they are partly flattened and only after the pulse has travelled some distance along the tube; and since the picture of the phenomenon as presented by the artery in situ is identical in every respect with that presented by the models there can be no doubt but that the phenomenon occurs in the artery in situ only when it, too, is partly flattened and over a length that is sufficient for its development. All of the pertinent observations we have made are in agreement with this conclusion. Thus the phenomenon is not present in the completely flattened artery; it appears only after a layer of blood remains in the artery at the close of diastole and develops or reaches its best development after the pulse has travelled some distance along the elliptical artery. Furthermore, the phenomenon disappears at a time when the behavior of the artery indicates that its walls are beginning to remain distended at the close of diastole. At such a time, of course, the artery would no longer be deformed by the compression. The change from the accelerating increase in diastolic residuum to the retarding increase unquestionably is indicative of this condition of the artery; and, as has been seen, the pre-anacrotic phenomenon then disappears. This deduction from the change in rate of the diastolic accumulation is supported by the observation that there is a similar tendency toward limitation of the rate of rise of the crest line early during decompression, which becomes obvious when the artery in systole has approximately the bore of the artery in diastole at the moment the retardation of the diastolic increase begins.

Therefore both indirect evidence and the experimental evidence derived from the study of models agree in indicating that the pre-anacrotic phenomenon disappears in arteries at the instant the compressing pressure falls below that which is necessary to produce elliptical deformation, in other words, at the moment the compressing pressure falls below the so-called diastolic pressure. The compressing pressure obtaining at the moment of disappearance of the pre-anacrotic phenomenon therefore must be a fairly accurate measure of the diastolic arterial pressure. Furthermore, the appearance of the phenomenon, while the compressing pressure is being lowered, indicates that the artery contains at the end of diastole a layer of blood that is deep enough to permit the phenomenon to develop. This will occur shortly after the compressing pressure falls below the so-called systolic pressure. We say "shortly after" because there are operative at this time a number of disturbing factors, to which reference has been made above, whose significance it is difficult to evaluate.

RELATION OF THE PRE-ANACROTIC PHENOMENON TO SOUND PRODUCTION

In models. The significance of the pre-anacrotic phenomenon in relation to sound production first became clear in the experiment with the rubber dam "artery" in which the records shown in figure 18 were made. It was found that when a pulse was made to travel through this "artery" containing water to a depth of 8 mm. a sound could be heard by means of a sphygmostethoscope so held that its button just touched the upper wall of the "artery." This sound differed very markedly in quality and intensity at different points along the course of the "artery." Close to the proximal end of the "artery" it was dull in quality and not particularly loud. Upon carrying the stethoscope down the tube in short steps, it was found that at 10 cm. the sound still was dull but somewhat louder. At 12 cm. it was still louder and had a snapping quality. At 15 cm. it still was snapping and louder. At 17 cm. it was much louder and ringing. At 20 cm. the sound had about the same quality and loudness as at 17 cm., but it was distinctly reduplicated. A little further along, the sound was still loud and snapping and more than one reduplication seemed to be audible. With further progress along the "artery" the sound became somewhat fainter though it was still snapping, and the reduplications were so frequent that the impression produced was that of a loud purr. At the far end of the "artery" the sound became louder again; but of this, more later.

Upon comparing the *form picture* of the "artery" with the *sound picture* it was found *a*, that the snapping quality first became evident (12 cm. down the tube) shortly after the negative wave of the pre-anacrotic phenomenon began to form (11 cm.); *b*, that the intensity of this sound increased as the pre-anacrotic waves became sharper; *c*, that the sound was reduplicated (at 20 cm.) shortly after a second negative wave began to form; and finally *d*, that the sound became a purr in the part of the "artery" through which a number of the smaller waves were running.

The same close correspondence between the snapping sound and the growth and decay of the sharp negative waves has obtained through all modifications of the conditions described above and in all types of "arteries." It is found furthermore, that the region of dull sounds extends down the "artery" as, with increasing fullness, the site of initiation of the pre-anacrotic phenomenon moves down; and that with the degree of fullness at which the pre-anacrotic phenomenon ceases to develop, the sound is dull along the entire length of the "artery."

Origin of the snapping sound. a. In tubes. To meet the possible objection that the sharp sounds are produced by a tapping of the stethoscope by the wall of the artery returning to its position against the stethoscope after the passage under it of the negative wave, the following experiment was performed. The "artery" was placed horizontally, either supported or suspended, about 1 cm. below the surface of water contained in an appropriate trough. These conditions, as has been seen, do not materially modify the pre-anacrotic phenomenon. A stethoscope (sphygmometroscope) was then arranged to move on guides which so held it that its button always just touched the surface of the water immediately over the "artery." As the stethoscope was moved along the length of the artery thus submerged the sequence of the sound changes remained the same as in the case of the "artery" in air, though the intensity of the sounds was somewhat diminished. In this experiment the "artery" at no time touches the stethoscope.

The close association of the snapping sounds with the pre-anacrotic waves points to, but does not prove, a causal relation between these two phenomena. In opening the discussion of this subject it is well to make it clear at once that the sound is not due to annular stretching of the wall of the "artery." This is proved by the observation that the pre-anacrotic phenomenon and the sharp sounds can develop in association with pulses which at no time are ample enough to put the wall of the "artery" on the stretch, at least in an annular direction.

In all cases the sense of touch clearly indicates that the snapping sound comes with the beginning of, or just before, the pulse; but simultaneous records of the pulse and of the sounds have not been made in the experiments with models. If fine sand is scattered along the rubber dam "artery" filled to a depth of 8 mm. and repeated pulses of increasing sharpness are sent through it, the sand is thrown off first at about 16 cm.; then between 11 and 19 cm.; and with more vigorous repetitions, between 11 and 30 cm. In addition there are indications that the "artery" tends to clear itself of sand in another region further down approximately at the point, namely 42 cm., where the negative wave reforms. The same phenomenon can readily be demonstrated by resting levers across the "artery."

These experiments show that the wall experiences the most vigorous shock about 16 cm. down the tube. It is in about this situation also that the loudest sound is heard (see above). In figure 18 it can be seen that in the vicinity of this point the anacrotic limb has encroached upon the negative wave to such an extent that the latter has been converted into a very brief, deep, sharp notch, and that this notch is preceded by a brief and small positive wave. At the same time the foot of the anacrotic limb has become very steep. At the point where the same picture returns the shock again increases in intensity; but it is not so strong as it was at 16 cm. presumably, because, as a result of friction, the pulse has lost some in force. These movements, the positive wave, the negative wave, the return of the anacrotic limb to the level of the base line are completed in a little over 0.01 second. This vibration rate in itself is fast enough to produce sound and probably is the cause of the sound, or at least initiates the sound. Results differing only quantitatively from the above can readily be obtained with the tire "artery."

b. In the artery in situ. If, now, we examine the records obtained from the artery in situ (figs. 3 and 4) with a view to determining the relation of the snapping sounds of Korotkoff to the pre-anacrotic phenomenon, it is seen that under the best of conditions the snapping sound beyond the compression chamber is first heard during decompression with about pulse *3*, or after a fall in the compressing pressure of about 4 mm. below the level at which the pulse first succeeds in reaching lever *3* and at a time the pre-anacrotic phenomenon already is fairly well developed. It should be recalled, however, that lever *3* is not quite at the lower orifice of the chamber and that the very first sounds may be at the limit of audibility. It is quite possible that the failure

of the sharp sound and of the pre-anacrotic phenomenon to develop in the same pulse is due to these conditions.

It is seen, furthermore, that under the best of conditions, that is, when the pre-anacrotic phenomenon disappears suddenly and the sounds are not masked by extraneous noises, the sound becomes dull with the very pulse in which the pre-anacrotic phenomenon disappears. When the pre-anacrotic phenomenon disappears gradually the agreement is not so close; but under these circumstances it seems probable that there is no sharp transition from snapping to dull sounds. All things considered, we feel justified in concluding that the pre-anacrotic phenomenon and the sharp sounds of Korotkoff are conterminal, and causally related.

Other possible causes of sound production during the critical stage of decompression are sudden annular stretching of the artery either by the pulse as it is propagated along the compressed segment or water hammer (see below) just below the compressed segment. There is, however, no very good evidence indicating that such stretching of the walls occurs through these actions as would be necessary for sound production. Thus at the time the sounds first are picked up the signs which we have taken as indicative of filling of the compressed segment by the pulse to its just round size usually are not yet in evidence even under lever *1*. Neither does the peripheral pulse at this time (see fig. 1, *a* in the paper referred to below) give any clear evidence of thetype of stretching of the artery we would expect to occur in association with water hammer.

Water hammer. In a preceding investigation of the mechanism of the Korotkoff sounds (3) we were led by our results to conclude that the arterial sounds are produced by the impact of the column of fluid, moving to fill the flattened artery, against the fluid filling the artery beyond the compressed segment with consequent stretching of the arterial wall there. None of the pertinent evidence presented in favor of that conclusion is opposed to the conclusion with regard to the cause of sound production reached in the present paper. Space prevents listing all of those observations here. The location of the "impact" under the arm band in man and the localization and time relation of sound production in the animal experiments alone will be referred to. The former was most distinctly felt under the lower part of the armlet; while the sounds seemed to originate close to the lower edge of the compression chamber, and were located by calculation early in the anacrotic limb of the pulse,—observations that are quite compatible

with the present point of view. On the other hand, the present series of experiments has brought to light one observation that is wholly incompatible with the water hammer hypothesis of sound production. In the experiments with the long models, the sharp sound originates at a point where a water hammer could not possibly develop. At the same time we still are of the opinion that where conditions favor the manifestation of water hammer it does become a factor in the production of sound. When the rubber tire model, in a partly flat state, is shortened so that the lower end becomes distended by the pulse, the sounds become much louder than in the long model as ordinarily used in our experiments. This possibly is due to water hammer. In animals, the addition to the anacrotic limb of the pulse, late in the critical stage of decompression, of what we have called the filling wave possibly is the graphic representation of what happens under these circumstances. But how much of this intensification is due to water hammer and how much to the energy liberated through reflection of the wave is not yet entirely clear.

Conduction of the pre-anacrotic phenomenon and of the sounds beyond the flattened segment. The routine method of examination for the Korotkoff sounds beyond the compressing armlet indicates that the sounds, and consequently the waves that produce them, are propagated down the artery. In a preceding paper (3) the movements of the artery *in situ* were recorded at the point where the sounds were picked up beyond the compressed segment. These records (see fig. 2 in that place) show, when the sounds are well developed, pre-anacrotic waves identical with those exhibited by the compressed segment as recorded in the present experiments.

Similar results have been obtained in experiments with the tire model by first increasing the pressure in the tube considerably above the level necessary to give to it a round form, and then flattening the proximal part by drawing a sheet of rubber dam over it. Under these circumstances the snapping sounds are well heard in the cylindrical tube beyond the flattened segment. They diminish in intensity, however, with the distance from the compressed segment.

SIGNIFICANCE OF THE SHARP SOUNDS AS BLOOD PRESSURE SIGNS

Inasmuch as the pre-anacrotic phenomenon and the sharp sounds are conterminal and inasmuch as the pre-anacrotic phenomenon begins practically at systolic compression, though this may depend somewhat

upon the length of the compressed segment, and ends presumably at diastolic compression, it follows that the first and the last of the sharp sounds also indicate the systolic pressure approximately, and the diastolic pressure exactly. As is well known there are some who still maintain that the disappearance of sound, the so-called fifth phase, rather than the beginning of the fourth phase, or dulling of sound, is the index to the diastolic pressure. Thus Hooker and Southworth (16), finding that in man the point of cessation of sound as determined by registration closely approximates the diastolic pressure, conclude that it serves all practical demands of clinical refinement as an index to the diastolic pressure. This may be the case in man under normal conditions. The fact, however, that in the dog and, under certain circumstances, in man, also, even the uncompressed artery emits a dull sound renders that view quite untenable. The present experiments conclusively prove that the dull sounds develop at the moment the artery remains round during diastole and therefore justify the conclusion that it is the appearance of the dull sounds and not the disappearance of all sound that indicates the diastolic pressure.

RATE OF PROPAGATION OF THE COMPRESSION PULSE

The rate of transmission of the pulse through the compression chamber increases during decompression. Though this increase is steady, it is most rapid in the earlier stages of decompression. Thus, the time required for the pulse to travel from lever *1* to lever *2*, when it first succeeds in reaching lever *2*, is from 33 (dog V) to 100 per cent (dog III) less than the time lost between the same levers at the moment the pulse first succeeds in reaching lever *3*.

At about the time the pre-anacrotic phenomenon disappears the rate is abruptly increased by an amount varying between 30 and even 100 per cent. Similar results were obtained in our earlier experiments (3) in which the compression oscillation in the chamber was recorded simultaneously with the pulse just peripheral to the chamber. In that paper the sudden increase in transmission time was taken to mean that the artery then remained filled to its just rounded size during diastole and that pulse propagation during the whole pulse cycle was therefore quickened by the tension of the artery which it then felt for the first time during the entire pulse cycle. While this undoubtedly is a factor, it is now evident that a part of this reduced transmission time is apparent rather than real, and is to be attributed to the dis-

appearance of the pre-anacrotic phenomenon. For, with this change the foot of the pulse wave as recorded by lever *3* (and also as recorded in the peripheral artery) presumably moves forward with respect to the pulse as a whole, thus apparently reducing the time interval between the arrival of the pulse at the upper and at the lower parts of the compressed artery.

SUMMARY AND CONCLUSIONS

1. Methods are described for recording photographically the movements of points on the exposed artery of the dog while and where it is being decompressed as in making blood pressure observations in man.

2. The records show that:

a. A thin layer of blood is pushed through the completely flattened segment shortly before the pulse succeeds in penetrating the full length of the segment.

b. This diastolic residuum for a time accumulates rather rapidly when the pulses begin to travel through the segment. The accumulation of the residuum is due to the fact that the time elapsing between pulses is insufficient for the compressing pressure to completely empty the segment of blood.

c. Shortly before the change of the third into the fourth phase sounds the diastolic residuum again accumulates rapidly and at an accelerating rate.

d. Beginning with the sounds of the fourth phase the accelerating accumulation of the diastolic residuum changes to a retarding accumulation. This inflection is taken to indicate the filling of the artery to the point where the walls remain stretched during diastole.

e. The pulse does not penetrate the full length of the segment when it starts to enter it; rather, under the conditions of the present experiments, the depth of penetration increases at this time 1.0 cm. for each 2.7 mm. Hg. of pressure decrement. The penetrativeness of the pulse seems to increase with the depth to which it penetrates. Attention is directed to the obvious error this behavior of the pulse, relative to the width of the armlet, introduces into all systolic readings of the blood pressure. The gradual penetration of the pulse during decompression explains the usual absence at this time of a *sudden* increase in amplitude of oscillations in oscillation records as obtained in observations on man, and suggests that beginning penetration of the pulse in the compressed

artery is indicated in oscillation records by a rapid, not necessarily a sudden, increase in the amplitude of the oscillations.

f. The first few pulses that penetrate may not distend the artery.

g. Variations in the radial amplitude of oscillation of the walls of the compressed segment indicate that the oscillatory index to the diastolic pressure is neither the first sudden diminution in amplitude nor the cessation of the abrupt diminution. Rather, it would seem that the index to the diastolic pressure lies between these changes, and where the accelerating diminution in oscillation amplitude begins to give way to a retarding diminution.

h. In the stage of decompression during which the artery is partly flat, which extends from the time when the pulse first begins to penetrate the length of the segment up to the time the fourth sound-phase begins, there is in evidence a process which is designated the *pre-anacrotic phenomenon:* the pulse front increases in steepness as it progresses through the flattened artery and small pre-anacrotic waves develop in front of it, the one immediately preceding the anacrotic limb usually being negative and undergoing an interesting metamorphosis. The process presumably resembles what has been termed by Lord Rayleigh the ripple phenomenon.

3. The pre-anacrotic phenomenon is displayed by partly flattened tubes with which it can be demonstrated that:

a. The wall of the tube experiences a sharp shock at the point where, during the progress of a pulse down the tube, the pre-anacrotic phenomenon becomes fully developed. Sand particles strewn on the tube are violently thrown from it at that point.

b. At the same point the sound produced by the artificial pulse, which more centrally is dull in quality, becomes sharp and snapping. The period of the pre-anacrotic waves here is short enough to make it possible to regard them as the source of the sound.

c. The sharp sounds and the pre-anacrotic phenomenon disappear at the same time, namely, when the tube is filled until round.

4. In the artery in situ the pre-anacrotic phenomenon and the sharp Korotkoff sounds also are conterminal and behave in every way as in the model.

5. It is concluded that the beginning of the first sound-phase, subject to certain errors, develops shortly after the compressing pressure falls below the systolic pressure; and that the change from the sounds of sharp quality to the sounds of dull quality fairly accurately indicates the moment the compressing pressure falls below the diastolic pressure.

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Supplementary note. In a supplementary note to a related subject,[2] Brooks and Luckhardt imply that I unjustifiably appropriated a method which they had demonstrated to me privately and that I hurried into print with the results obtained through its use in order to forestall theirs.

Lest the unanswered presence of this note in the pages of the Journal, where it will stand for all time, give the impression that it had editorial sanction, which it did not, and that its implications are justifiable, I reluctantly, even at this late date, use this opportunity, granted at the time by the Editor, to present the facts.

Freed entirely of implication they are these:

1. The method was not demonstrated to me in private. It was demonstrated publicly at a public meeting of the American Physiological Society.

2. I was not present at the demonstration; therefore, as subsequently became evident, the method actually employed by me happened to differ quite materially from theirs.

3. Brooks and Luckhardt in my paper are given full and unqualified credit for the method which I used under the impression that it was theirs, and that the public demonstration of a method is for the purpose of making that method available for the furtherance of science.

4. My paper did not appear until more than fourteen months had elapsed after their public demonstration.

5. Inasmuch as I was working at my own problems, my method of handling the subject was altogether different from theirs; so that

6. Instead of arriving at the same conclusions, in the few instances in which they touch upon the same subjects, our conclusions often are quite at variance.

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[2] This Journal, 1916, xl, 49.